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VOLUME 29 DECEMBER, 1951 NUMBER 12

THE BIOGENESIS OF ALKALOIDS

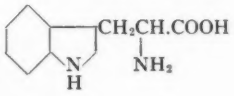
IV. THE FORMATION OF GRAMINE FROM TRYPTOPHAN IN BARLEY[1]

BY K. BOWDEN[2] AND LÉO MARION

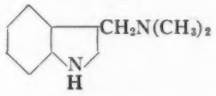
Abstract

dl-Tryptophan-β-C^{14} was fed to sprouting barley and radioactive gramine isolated from the shoots. Activity in the alkaloid resided entirely in one position, corresponding to that in the tracer administered to the plants, indicating that tryptophan is a precursor of gramine.

The alkaloid gramine, occurring in barley (4–8) and the Asiatic sedge *Arundo donax* L. (12), is a member of the indole group of alkaloids which includes abrine and hypaphorine, both of which are N-methyl derivatives of tryptophan. In view of the wide occurrence of tryptophan in the plant world and its close relationship to abrine and hypaphorine it seems very probable that it is a precursor of both these alkaloids. The same connection between tryptophan and gramine, first suggested by von Euler (8), is not as certain, as it would involve a complicated change of the side chain.



Tryptophan



Gramine

In order to examine this possibility we have administered tryptophan, labelled with C^{14} in the β-position, to barley and degraded the alkaloid formed. Barley is suitable for tracer experiments as it can be grown in glass dishes and requires only the addition of water. In addition, certain types such as Charlottetown No. 80 quickly accumulate the alkaloid in the leaves in concentrations which permit its isolation without much difficulty.

To avoid undue dispersion of radioactivity caused by metabolism the tracer was fed to the plants on the 6th day of sprouting and the leaves were harvested on the 11th day.

The alkaloid isolated from the leaves remained radioactive after repeated purification indicating that part of the tryptophan molecule has been utilized

[1] *Manuscript received August 29, 1951.*
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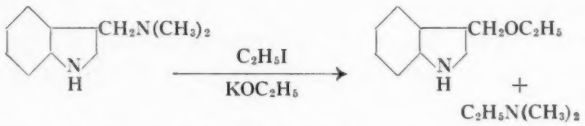
[2] *National Research Council of Canada Postdoctorate Fellow (1950-51).*

in the formation of the gramine. It was diluted with inactive gramine and subjected to two series of degradations in order to determine the location of the radioactive atom or atoms in the molecule.

(*a*) Madinaveitia (11) has shown that gramine will undergo the following reaction:



Degradation of the labelled alkaloid by this means yielded inactive ethyldimethylamine (picrate) and 3-ethoxymethylindole which had a molecular activity equal to that of the starting material, showing that the dimethylamino group in the original alkaloid contained no labelled atoms (Table I).

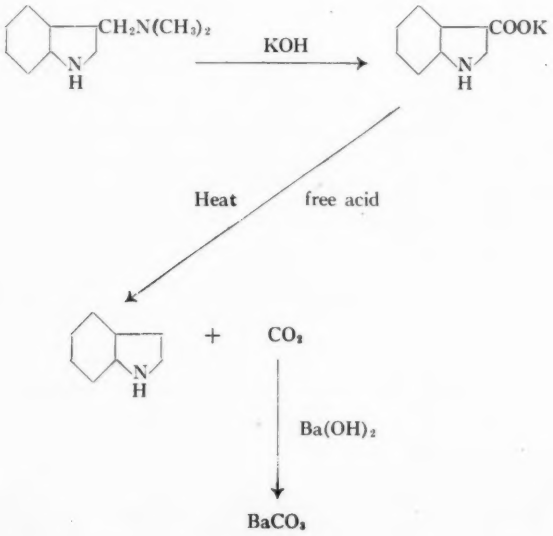
TABLE I

ACTIVITIES OF GRAMINE AND DEGRADATION PRODUCTS

Compound	Disintegrations per minute per millimole
Gramine	3.06×10^4
Ethyldimethylamine picrate	0
3-Ethoxymethylindole	3.08×10^4

(*b*) It was found that fusion of gramine with potassium hydroxide gave the potassium salt of indole-3-carboxylic acid. The free acid yielded indole and carbon dioxide on heating and the latter was converted into barium carbonate by absorption in barium hydroxide solution.

i.e.



In order to test for possible rearrangements which might occur under the rather drastic conditions of fusion with potassium hydroxide, a synthetic sample of gramine with C^{14} in the side chain was submitted to the same treatment. The activities of the products from this material and from the natural alkaloid are given in Table II.

TABLE II

ACTIVITIES OF DEGRADATION PRODUCTS FROM NATURALLY AND SYNTHETICALLY LABELLED GRAMINE

Compound	Disintegrations per minute per millimole	
	Natural	Synthetic
Gramine	3.06×10^4	2.47×10^4
Indole-3-carboxylic acid	3.03×10^4	2.48×10^4
S-Benzyl-iso-thiouronium salt of indole-3-carboxylic acid	3.05×10^4	2.39×10^4
Indole picrate	0	0
Barium carbonate	3.04×10^4	2.46×10^4

The activities of the products obtained from the fusion of synthetic gramine with potassium hydroxide show that no rearrangements occur. The results obtained from the degradation of the natural gramine indicate that only one atom in the alkaloid molecule was labelled. The position of this labelled atom in the alkaloid corresponds with that of the C^{14} in the tryptophan fed to the plants, strongly suggesting that tryptophan is a precursor of gramine in barley.

Experimental

(All radioactive compounds were measured as thin samples in a Radiation Counters Laboratory "Nucleometer" and appropriate corrections made.)

Preparation of Labelled Tryptophan

dl-Tryptophan-β-C^{14} was prepared by suitable modifications of known procedures (1, 9, 13) using one-ninth of a millicurie of C^{14}-formaldehyde.

Administration of Tracer to Barley

A 720 gm. sample of Charlottetown No. 80 barley was grown as previously described (10), in 12 Pyrex dishes and was watered each day with distilled water, using an atomizer. The trays were stored in a closed germination cabinet.

On the 6th day of sprouting, *dl*-tryptophan acetate (0.501 gm.) containing one molecule of crystallization of acetic acid, and with an activity of 4.1×10^7 disintegrations per minute per millimole (total activity = 6.36×10^7 disintegrations per minute) was fed to the barley in 600 ml. of distilled water.

Isolation of Labelled Gramine from Barley

On the 11th day of sprouting the shoots were harvested by clipping off as close as possible to the root mat and were dried in an oven at 60°. The dry

shoots weighed 80 gm. They were extracted without further treatment with methyl alcohol in a Soxhlet extractor for 48 hr. and the extract then evaporated down to dryness under reduced pressure.

The residue was taken up in 2*N* sulphuric acid (200 ml.) and then extracted twice with 100 ml., then four times with 50 ml. of diethyl ether. The aqueous layer was cooled in ice, and sodium hydroxide (25 gm.) in a minimum of water was added with stirring. The mixture was extracted three times with 100 ml., then four times with 50 ml. of diethyl ether and the ethereal solution washed with water (100 ml.).

The ethereal solution was dried over anhydrous sodium sulphate and the solvent removed under reduced pressure to give a crystalline residue (0.45 gm.) which distilled under reduced pressure as a liquid (b.p. 90–100° at 10^{-4} mm.) and which crystallized in the receiver (0.28 gm.). The product was recrystallized from benzene to give long needles (0.2456 gm.), m.p. 133° undepressed on admixture with an authentic specimen of gramine.

Anal.	Calcd. for $C_{11}H_{14}N_2$:	C, 75.84; H, 8.10; N, 16.09
	Found:	C, 76.01; H, 8.19; N, 16.06%.

The sample had specific activity of 583.9 disintegrations per minute per milligram. It was recrystallized from benzene and 0.2155 gm. of the product mixed with 0.4995 gm. of inactive gramine and the whole recrystallized from benzene to give 0.6574 gm. of gramine with specific activity of 3.06×10^4 disintegrations per minute per millimole. Degradation studies were carried out on this material.

Degradations of Gramine Obtained from Plant

(*a*) The diluted gramine (0.2 gm.) was dissolved in absolute alcohol (1 ml.) and freshly distilled ethyl iodide (0.2 ml.) added. Crystals of the ethiodide began to form almost immediately. After two hours at room temperature ethanolic potassium hydroxide (0.5 ml. of 25% solution) was added and the air was slowly passed through the mixture for seven hours. The issuing gases were bubbled through a saturated solution of picric acid in methyl alcohol.

The resulting ethyldimethylamine picrate was filtered off and dried (0.0777 gm.) and after recrystallization from methyl alcohol melted at 204–205°. Water was added to the reaction mixture, giving an oil which crystallized. This was filtered off, taken up in ether, dried, and after removal of the solvent, recrystallized from petroleum (b.p. 50–80°) as needles (0.112 gm.), m.p. 67.5°.

Anal.	Calcd. for $C_{11}H_{13}ON$:	C, 75.37; H, 7.47; N, 8.0
	Found:	C, 75.29; H, 7.24; N, 7.89%.

Madinaveitia (11) gives the melting point of 3-exthoxymethylindole as 93–94°. We were unable to confirm this, our product always melting at 67.5°. This was also the melting point of the product when methyl iodide,

in ethyl alcohol, was used in place of ethyl iodide, indicating the source of the ethyl group to be the ethyl alcohol.

The activities of the compounds in the above degradation are given in Table I.

(*b*) Gramine obtained from the plant and synthetic labelled gramine were both degraded by the following method, the activities of the resulting compounds being given in Table II.

Gramine (0.4 gm.) was added in small portions to potassium hydroxide pellets (5 gm.), heated just to fusion in a platinum crucible. When the reaction was complete and the mixture had cooled, it was taken up in a little distilled water and then extracted twice with an equal volume of diethyl ether. The aqueous portion was cooled in ice and acidified with concentrated sulphuric acid, extracted well with diethyl ether, and the ethereal solution dried over anhydrous sodium sulphate. On removal of the solvent under reduced pressure, crude indole-3-carboxylic acid was obtained as a pale yellow solid, m.p. 195° (decomp.) (0.2916 gm.). After being washed with a little diethyl ether it melted at 209° (decomp.) (0.2112 gm.). The free acid readily decomposes on heating in the solid state or in solution.

The S-*benzyl-iso-thiouronium* salt crystallized from water, m.p. 157.5° (decomp.).

Anal. $C_{17}H_{17}O_2N_3S$ requires C, 62.38; H, 5.21; N, 12.83
Found: C, 62.49; H, 5.08; N, 12.80%.

It showed no depression of melting point when mixed with the corresponding salt of an authentic specimen of indole-3-carboxylic acid prepared from indole through the aldehyde (2, 3).

Indole-3-carboxylic acid, from the potassium hydroxide fusion (0.104 gm.) was decarboxylated in a small vertical tube fitted with a nitrogen inlet leading almost down to the bottom of the tube. The issuing gases were bubbled through barium hydroxide solution (5 ml. of 4%). The apparatus was first flushed out with carbon dioxide-free nitrogen and the lower part of the tube containing the indole carboxylic acid was slowly heated in a metal bath to 190°, at which temperature evolution of carbon dioxide started. Decarboxylation was complete at 220° and the temperature was finally raised to 240°, when indole distilled up the tube and crystallized out on the upper parts. Residual carbon dioxide was swept out of the tube with a stream of nitrogen. The precipitated barium carbonate was filtered off, washed well, and dried (0.0918 gm.).

The indole (0.0503 gm.) was converted into its picrate which crystallized from methyl alcohol as crimson plates and melted at 187.5°, undepressed on admixture with an authentic specimen of indole picrate.

Anal. Calcd. for $C_{14}H_{10}N_4O_7$: C, 48.54; H, 2.91; N, 16.17
Found: C, 48.78; H, 2.77; N, 15.86%.

Acknowledgment

The authors are indebted to Mr. R. B. MacLaren of the Experimental Station, Charlottetown, P.E.I., for supplying the barley used in these experiments.

References

1. ALBERTSON, N. F., ARCHER, S., and SUTER, C. M. J. Am. Chem. Soc. 67: 36. 1935.
2. BOYD, W. J. and ROBSON, W. Biochem. J. 29: 555. 1935.
3. ELLINGER, A. Ber. 39: 2515. 1906.
4. EULER, H. v. and ERDTMAN, H. Ber. 69: 743. 1936.
5. EULER, H. v. and HELLSTRÖM, H. Z. physiol. Chem. 208: 43. 1932.
6. EULER, H. v. and HELLSTRÖM, H. Z. physiol. Chem. 217: 23. 1933.
7. EULER, H. v., HELLSTRÖM, H., and HAGEN, J. Arkiv Kemi, Mineral. Geol. 11B, No. 36. 1934.
8. EULER, H. v., HELLSTRÖM, H., and LÖFGREN, N. Z. physiol. Chem. 234: 151. 1935.
9. HEIDELBERGER, C. J. Biol. Chem. 179: 139. 1949.
10. KIRKWOOD, S. and MARION, L. Can. J. Chem. 29: 30. 1951.
11. MADINAVEITIA, J. J. Chem. Soc. 1927. 1937.
12. OREKHOV, A., NORKINA, S., and MAXIMOVA, T. Ber. 68: 436. 1935.
13. SNYDER, H. R. and SMITH, C. W. J. Am. Chem. Soc. 66: 350. 1944.

THE BIOGENESIS OF ALKALOIDS

V. RADIOAUTOGRAPHS OF BARLEY LEAVES FED WITH TRYPTOPHAN-β-C^{14} [1]

By K. Bowden[2] and Léo Marion

Abstract

Radioautographs of leaves of barley fed with C^{14}-tryptophan have been taken at intervals. In the early stages of development a radioactive area appeared at the bottom of the leaves, which was due mainly to free tryptophan. Later an area, consisting mainly of gramine, developed at the top of the leaves.

In a previous publication (2) we have shown that Charlottetown No. 80 barley fed with tryptophan-β-C^{14} produces in the leaves gramine, labelled in the same position in the side chain, indicating that the amino-acid is a precursor of the alkaloid.

In order to study the assimilation of tryptophan by the plant and the accumulation in it of the alkaloid, we have administered the labelled amino-acid to the plant and have taken radioautographs of the leaves harvested at intervals.

Approximately sixty seeds of Charlottetown No. 80 barley were grown in a Petri dish in a closed germination cabinet and were moistened each day with distilled water. On the fifth day of sprouting the acetate of *dl*-tryptophan-β-C^{14}(0.1 gm.), with an activity of 4.1×10^7 disintegrations per minute per millimole was fed to the plants in distilled water (30 ml.).

Twenty-four hours afterwards and each day following, for five days, seven leaves were harvested by clipping them off as close as possible to the seeds and were dried flat between glass slides (2 in. $\times$ 7 in. $\times$ $\frac{1}{4}$ in.) in an oven at 65°. In order to obtain some measure of the rate of absorption of the amino-acid by the plant and the exposure to the X-ray film that each leaf would require to give a satisfactory radioautograph, two dried leaves were assayed from each day's harvest. The leaves, cut into suitable portions, were pressed flat on to the sticky surface of cellophane tape covering a small cardboard disk. The leaf-covered disk was then assayed in a Radiation Counters Laboratory "Nucleometer". The "count", without any corrections for sample thickness, geometry, etc., for leaves harvested 24 hr. after the administration of the tracer was 409 per leaf per minute, indicating that entry of the amino acid into the plant was fairly rapid. The count rose to 2000 per leaf per minute for leaves harvested subsequently.

Axelrod (1) has found that about ten million β-particles per square centimeter produce a satisfactory radioautograph. As the total activity of a leaf

[1] *Manuscript received August 29, 1951.*
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[2] *National Research Council of Canada Postdoctorate Fellow (1950-1951).*

gives only a rough indication of the exposure to X-ray film required, five leaves from each day's harvest were set up in separate radioautograph units and were developed after varying lengths of exposure.

Each unit consisted of a strip of Kodax "No Screen" X-ray film (7 in. × 2 in.) against which the flat leaf was placed. The two were sandwiched between glass slides (7 in. × 2 in. × $\frac{1}{4}$ in.) held together by metal clips along the long edges. Each unit was wrapped in light-proof paper and stored in a refrigerator until developed.

Development was carried out in Ansco "Liquadol" X-ray developer for three minutes at 20° and the films were fixed for five minutes at 20° in Ansco "Liquafix" acid fixer. All manipulations of the X-ray film were carried out by the light of a Kodax 6B safelight.

A series of radioautographs, each of one week's exposure to X-ray film, are given in Plate I. The first is of a leaf harvested one day after the tracer was fed to the plants and those which follow are of leaves harvested at daily intervals from then onwards.

Discussion

As may be seen from the radioautographs, an active area appears at the bottom of the leaf on the first day and slowly extends upwards on the second and third days. On the fourth day an active area makes its appearance at the top of the leaf and this area of concentration increases on the fifth and sixth days.

It has been shown by ultraviolet absorption studies that the concentration of gramine in a barley leaf increases towards the tip of the leaf (3). It seemed to us likely, therefore, that the area of activity in the lower part of the leaf might be due to tryptophan or a gramine precursor, whilst the area at the tips of the leaves might be due to an accumulation of gramine.

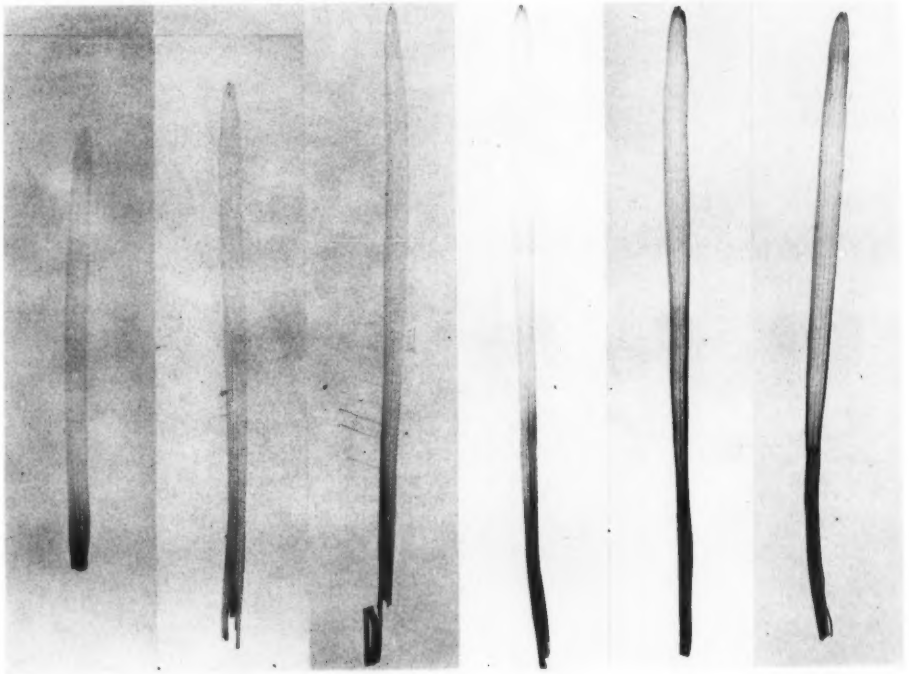
Although tryptophan occurs widely in plants and has been isolated from barley protein (4) it could not merely be assumed to exist in a simple form in the lower area of the leaves of Charlottetown No. 80 barley.

In order to test whether this was so, a dried radioactive leaf, harvested six days after the administration of the tracer, was cut transversely, separating from each other the two areas of radioactivity shown by radioautography. The two areas were then assayed separately in the "Nucleometer".

The top section of the leaf was cut into portions and soaked overnight in methyl alcohol (5 ml.) containing inactive gramine (25 mgm.). The solution was then filtered and the alcohol removed under reduced pressure and the gramine recrystallized from benzene to constant activity.

The lower portion of the leaf was treated similarly with a solution of inactive tryptophan acetate (25 mgm.) in glacial acetic acid (2.5 ml.). After filtration

PLATE I



I II III IV V VI

of the solution, the tryptophan was recrystallized to constant activity from glacial acetic acid.

By this means it was possible to show that approximately 58% of the activity in the area at the top of the leaf was due to free gramine, whilst approximately 77% of the activity in the lower part of the leaf was due to free tryptophan, thus confirming that the radioactive areas were mainly concentrations of gramine and its probable precursor, tryptophan, respectively.

Although these figures are only approximate, the lower proportion of gramine extracted in comparison to the tryptophan is probably significant, suggesting the presence of metabolites closely related to gramine in the area of activity at the top of the leaf.

By feeding labelled gramine (as acetate) to the barley, radioautographs were obtained showing the active areas only at the bottom of the leaves, indicating that gramine does not tend to travel to the top of the leaf and accumulate there. It would appear then that the active area which appears at the top of the leaf when the plant is fed with labelled tryptophan is the site of formation of the gramine and not just an area of accumulation of the alkaloid.

Our experiments show that tryptophan is readily absorbed by the plant and can exist as such in the leaves and also that gramine is formed in the leaves at least up to the 11th day of growth. It has been shown by spectroscopic means that during this period the total amount of gramine in the leaves remains constant (3). Therefore it appears probable that during this period the rates of formation and utilization of the gramine in the plant are approximately equal. After one month the gramine disappears from the leaf (3) indicating that in later stages of development of the plant this relationship no longer holds.

Acknowledgments

The authors are indebted to Prof. L. F. Bélanger of Ottawa University and to Dr. A. Morrison of the Radiology Department of the National Research Council, for advice and materials.

References

1. AXELROD, D. J. U.S.A.E.C., Univ. of California, BP-111.
2. BOWDEN, K. and MARION, L. Can. J. Chem. 29: 1037. 1951.
3. BRANDT, K., EULER, H. VON, HELLSTRÖM, H., and LÖFGREN, N. Z. physiol. Chem. 235: 37. 1935.
4. TILLMANS, J. and ALT, A. Biochem. Z. 164: 135. 1925.

CINÉTIQUE DE L'OXYDATION DE LA FORMALDÉHYDE PAR L'EAU OXYGÉNÉE EN SOLUTION ALCALINE[1]

PAR JEAN-B. JAILLET[2] ET CYRIAS OUELLET

Résumé

L'oxydation de la formaldéhyde a été étudiée, par mesure de l'hydrogène dégagé, dans des solutions aqueuses de pH 12-13, entre 0° et 35°C. et à des concentrations de 0.05 à 0.4 mole/litre. La réaction est du second ordre par rapport au temps, mais le taux varie par un facteur de 30 suivant les concentrations et proportions initiales des réactifs. L'énergie d'activation est de 14.5 kcal./mole. L'addition d'hydroquinone ou de sulfate de sodium ou un accroissement modéré de la surface de Pyrex sont sans influence. La vitesse de décomposition du peroxyde de diméthylol est identique à celle du mélange formaldéhyde—eau oxygénée équivalent. Le delta cryoscopique des solutions passe par un minimum. On examine l'hypothèse de la décomposition bimoléculaire d'un peroxyde intermédiaire et de l'intervention de polymères de la formaldéhyde.

Introduction

En solution aqueuse fortement alcaline, le réaction de l'eau oxygénée avec la formaldéhyde donne lieu à un dégagement quantitatif d'hydrogène suivant l'équation:

$$2HCHO + H_2O_2 + 2NaOH = 2HCOONa + 2H_2O + H_2.$$

Cette réaction, découverte par Blank et Finkenbeiner (3), a été utilisée pour le dosage gazométrique de la formaldéhyde. L'oxygène et l'acide carbonique peuvent aussi se former dans certaines conditions (1, 7, 8, 11, 14, 19), mais nous avons opéré en solutions diluées et fortement alcalines (pH 13) où l'on obtient seulement l'acide formique et l'hydrogène. On obtient aussi ces deux produits en remplaçant l'eau oxygénée par l'oxyde cuivreux (10) ou par une oxydation anodique (15). Glasstone et Hickling (9) attribuent ce dernier effet à l'eau oxygénée formée par la recombinaison des radicaux hydroxyles provenant de la décharge des ions hydroxyles à l'anode. L'oxydation de la vanilline (16) semble être le seul autre cas connu de production d'hydrogène par oxydation d'une aldéhyde au moyen de l'eau oxygénée.

Dans le but d'élucider le mécanisme de la réaction entre l'eau oxygénée et la formaldéhyde, Fry et Payne (7) ont étudié les produits obtenus dans diverses conditions. Wirtz et Bonhoeffer (22) ont montré, à l'aide du peroxyde de deutérium, que l'oxygène dégagé provient uniquement de la formaldéhyde, tel que prévu par la théorie de la déshydrogénation de Wieland (20, 21). Du point de vue cinétique, Kastle et Loevenhart (12) ont calculé des constantes du second order à 50, 60 et 70°C., Hatcher et Holden (11) ont suivi la neutralisation de la soude par l'acide formique produit. Pour compléter ces données, nous avons étudié la cinétique de la réaction dans les conditions où elle obéit à l'équation chimique donnée ci-dessus.

[1] *Manuscrit original reçu le 20 décembre 1950, et sous forme revisée le 5 septembre, 1951. Contribution du Département de Chimie, Université Laval, Québec. P.Q.*

[2] *Boursier Cominco. Adresse actuelle: Ecole Polytechnique, Montréal, P.Q.*

Méthode experimentale

Pour suivre la marche de la réaction, on mesurait la pression de l'hydrogène dégagé au moyen d'un manomètre différentiel à mercure. Dans le but de prélever, pour fins d'analyse, plusieurs échantillons de gaz au cours d'une même réaction, on s'est servi d'un appareil de grand volume (1600 cc.), qui se composait d'un ballon en Pyrex contenant un agitateur magnétique et relié aux autres organes au moyen d'un réfrigérant à reflux qui empêchait la distillation de la vapeur d'eau. Après avoir introduit la solution alcaline de formaldéhyde, on évacuait le système au moyen d'une trompe à eau puis on injectait rapidement une quantité mesurée de solution d'eau oxygénée au moyen d'une burette soudée au col du ballon et dont l'orifice était un long tube capillaire. Pour empêcher la solution de s'échauffer au début des réactions les plus rapides, on ajoutait graduellement par le capillaire une quantité connue (10-20 cc.) d'eau glacée; dans le cas des réactions à 0°C., on incorporait à la solution de formaldéhyde 10 grammes de glace pulvérisée. La réaction terminée, on aspirait un échantillon du gaz dans une ampoule évacuée préalablement au moyen d'une pompe Hyvac et on refoulait cet échantillon dans un analyseur à gaz de Haldane, modèle C, comportant une burette de 21 cc. dont 6 cc. sont gradués au centième de cc.

Une solution fraîche de soude et de formaldéhyde était préparée avant chaque expérience en dissolvant dans une solution de soude fraîchement titrée une quantité pesée à 0.001 gramme près de paraformaldéhyde de marque Eastman, qui se dépolymérise rapidement en milieu alcalin pour donner de la formaldéhyde monomérique. Des analyses de la paraformaldéhyde, effectuées au moyen de la réaction même que nous voulions étudier, ont indiqué une teneur de 99.2 à 99.7% en formaldéhyde. Les solutions d'eau oxygénée étaient titrées au permanganate après avoir été préparées par dilution soit de solutions à 90% de Buffalo Electrochemical Co., soit d'une solution à 30% de McArthur Chemical Co. On a obtenu des vitesses de réaction identiques avec des solutions d'eau oxygénée contenant des stabilisateurs ou n'en contenant pas.

Résultats

Observations préliminaires

Pour des motifs de commodité, la plupart des réactions ont été effectuées dans des conditions telles que le volume d'hydrogène dégagé fût de l'ordre de 500 cc. ramené aux conditions normales (T.P.N.). Sauf indication contraire, le volume de la solution était de 450 cc. Les quantités de réactifs employées dans les divers mélanges étaient généralement des multiples simples d'une quantité unitaire de 0.0225 mole, correspondant à une concentration de 0.05 mole par litre. Pour faciliter les références aux diverses réactions, les trois réactifs sont partout désignés dans l'ordre: formaldéhyde-eau oxygénée-soude (F-P-S) et leurs concentrations indiquées par trois chiffres placés dans le même ordre. Par exemple, la cote 4-1-2 représente un mélange de 0.2 mole/litre de HCHO, 0.05 mole/litre de H_2O_2, 0.1 mole/litre de NaOH. Le chiffre 1 représente 0.05 mole/litre.

TABLEAU I

RENDEMENTS DE DIVERSES RÉACTIONS EN CC. DE H_2 RAMENÉS À 0°C. ET 760 MM. (T.P.N.)

Solution F-P-S	H_2 cc. T.P.N.	$\frac{H_2 \text{ obs.}}{H_2 \text{ calc.}}$	Solution F-P-S	H_2 cc. T.P.N.	$\frac{H_2 \text{ obs.}}{H_2 \text{ calc.}}$	O_2 %
2-1-2	492	0.98	1-1-2	263	1.05	–
2-1-8	508	1.01	2-2-2	355	0.71	–
6-1-2	498	1.00	8-1-2	485	0.97	–
4-1-2	485	0.97	16-1-2	466	0.93	3.5
			20-1-2	407	0.81	9
10-1-2	490	0.98	24-1-2	365	0.73	6
6-1-2	492	0.98	24-1-0	–	–	1
$8\text{–}\frac{3}{4}\text{–}2$	393	1.05	0-2-2	–	–	1

Des mesures préliminaires ont vérifié que la quantité d'hydrogène dégagée était, dans presque tous les cas, celle que prévoit l'équation:

$$2HCHO + H_2O_2 + 2NaOH = 2HCOONa + 2H_2O + H_2$$

On voit, par les exemples donnés au tableau I, comment cette quantité est limitée par celle de l'un des réactifs. On y trouve aussi les rares exceptions où le rendement est inférieur au rendement théorique.

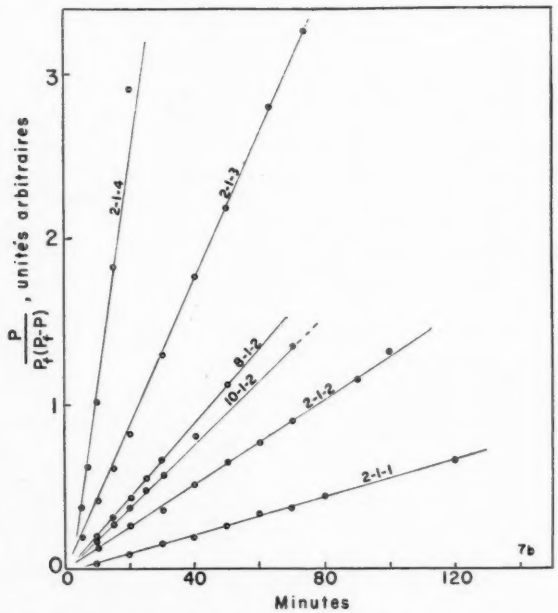


FIG. 1. *Série de réactions à 0°C. dans 450 cc. de solution, montrant la variation linéaire en fonction du temps de l'expression*

$$\frac{P}{P_f(P_f - P)}.$$

Trois solutions, contenant chacune de la formaldéhyde en grand excès, ont fourni des quantités d'oxygène appréciables, pas suffisantes cependant pour combler le déficit en hydrogène. Afin de voir si cet oxygène devait être attribué à la simple décomposition de l'eau oxygénée durant ces réactions très lentes, des mesures ont été effectuées sur des solutions d'eau oxygénée avec de la formaldéhyde seulement (24-1-0) ou de la soude seulement (0-2-2). Les quantités d'oxygène recueillies au bout de trois et une heures respectivement sont très faibles. Il semble donc que l'oxygène dégagé en présence d'un grand excès de formaldéhyde soit véritablement un produit latéral de la réaction d'oxydation.

La recherche de l'oxyde de carbone au moyen de papiers imbibés de chlorure de palladium a donné des résultats négatifs dans tous les cas. Il en fut de même

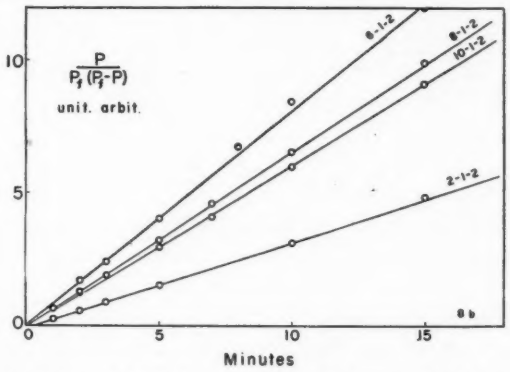


FIG. 2. *Série de réactions à 25°C. dans 450 cc. de solution, montrant la variation linéaire en fonction du temps de l'expression*

$$\frac{P}{P_f(P_f - P)}.$$

de celle de l'anhydride carbonique, soit par l'hydroxyde de baryum dans la solution après la réaction, soit par la potasse dans l'appareil de Haldane.

A la suite de ces constatations, nous nous sommes contentés d'analyser le gaz chaque fois que le rendement s'écartait de plus de 2% du rendement théorique. Le prélèvement d'échantillons de gaz au cours de la réaction est devenu, à plus forte raison, superflu. Les trois réactions indiquées au tableau I restent les seules qui aient produit de l'oxygène.

Les courbes de la figure 8 indiquent l'allure générale de la réaction. Les vitesses sont reproductibles avec une précision de l'ordre de 2%, à condition que le taux d'agitation soit supérieur à un taux minimum qui fut déterminé dans des essais préliminaires.

L'addition de sulfate de sodium à la concentration de 0.5 mole/litre et celle d'hydroquinone à la concentration de 0.001 mole/litre ont donné des courbes superposables à celles obtenues avec les mêmes solutions sans ces additions.

L'étape qui limite la vitesse n'est donc pas de nature ionique et ne comporte probablement pas de chaînes. De même, l'addition de Pyrex pulvérisé présentant une surface de l'ordre de deux fois celle du ballon n'a produit aucun effet. Cependant, l'addition de 64 grammes de Pyrex pulvérisé, dont nous avons estimé la surface à 8000 cm.^{2}, a modifié radicalement l'allure de la réaction. Cette réaction est plus lente au début que la réaction normale, mais sa vitesse reste constante (ordre zéro) et elle parvient à terme avant l'autre. Le produit de cette réaction était normal, soit 99% du rendement théorique en hydrogène. Cet effet remarquable mériterait d'être étudié en détail. On peut cependant conclure que la réaction ordinaire, dans les conditions où nous l'avons étudiée, est homogène.

Ordre de la réaction

L'analyse des courbes a révélé qu'elles n'obéissent à aucun ordre simple si l'on introduit dans les équations les valeurs des concentrations initiales des réactifs et celles des concentrations de l'hydrogène produit. Par contre, toutes les réactions, à de très rares exceptions près, suivent avec une assez bonne approximation la relation (1*a*)

$$A = \frac{1}{t} \cdot \frac{P}{P_f(P_f - P)} \quad (1a); \quad k_2 = \frac{1}{t} \cdot \frac{x}{a(a-x)} \quad (1b)$$

où P est la pression de l'hydrogène au temps t, P_f la pression finale et A une constante. Les droites des figures 1 et 2 illustrent cette relation par deux séries d'exemples.

La forme de (1*a*) est identique à celle de (1*b*), équation du second ordre pour le cas spécial où les deux réactifs sont à des concentrations initiales égales, soit $a = b$. Ici, la pression P de l'hydrogène représente la concentration x d'un produit de la réaction, tandis que la concentration initiale a des réactifs correspond à la concentration finale de l'hydrogène, proportionnelle à P_f. La constante A est l'analogue d'une constante du second ordre k_2. Tout indique que la première étape de la réaction est la formation rapide et probablement irréversible d'un complexe, dont la quantité est forcément limitée par celle de l'un des réactifs, et qui se décompose ensuite suivant une loi du second ordre.

Quelques mesures de la variation du pH en fonction du temps, effectuées au moyen d'une électrode de verre sur des mélanges 2-1-2 et 2-1-1 à 25°C., indiquent que la concentration des ions H^+ croît suivant une loi du second ordre. Mais ces résultats, obtenus sans précaution spéciale et en présence du système oxydo-réducteur formaldéhyde-eau oxygénée, ne constituent qu'une confirmation semi-quantitative de l'ordre de la réaction.

Influence des concentrations

Le comportement de la réaction se complique de façon inattendue lorsque l'on fait varier les concentrations et les proportions initiales des réactifs. A diverses concentrations d'une solution dans laquelle la formaldéhyde, l'eau oxygénée et la soude se trouvent dans les proportions 4-1-4, la vitesse initiale v_i,

exprimée en cm. Hg par minute, varie sensiblement comme le carré de la concentration (Tableau II), comme on devait s'y attendre. Cependant le coefficient A de l'équation (1a), loin de rester constant, varie à peu près comme v_i.

TABLEAU II

VARIATION DE LA VITESSE INITIALE v_i ET DU COEFFICIENT A À 25°C., EN FONCTION DE LA CONCENTRATION C_F EN MOLES PAR LITRE DE LA FORMALDÉHYDE POUR DES MÉLANGES DE COMPOSITION 4-1-4

C_F moles/litre	0.125	0.16	0.23	0.30	0.45	0.83
v_i	1.5	2.6	5.5	8.8	17.9	–
v_i / C_F^2	95	105	95	105	85	–
A	0.224	–	0.79	–	3.53	14.1
A / C_F^2	14.5	–	15	–	17	20

En appelant $\left(\frac{dP}{dt}\right)_i$ le taux initial de dégagement de l'hydrogène, on peut donc écrire:

$$v_i = \left(\frac{dP}{dt}\right)_i = A(P_f - P)^2 \qquad (1c)$$

Mais nous allons voir que A, qui joue le rôle d'une constante du deuxième ordre pour chaque réaction individuelle, dépend du rapport initial des concen-

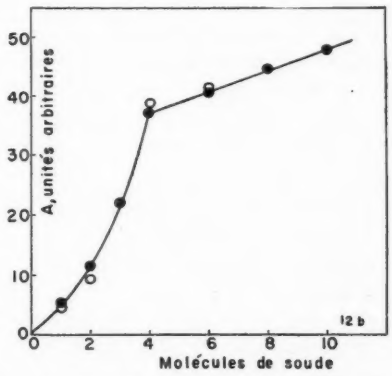


FIG. 3. *Influence de la proportion de la soude sur la vitesse représentée par le facteur A (équation 1a). Réactions à 0°C. dans 450 cc. (cercles noirs) et 210 cc. (cercles blancs), valeurs de A réduites proportionnellement pour montrer la coincidence des courbes. En abcisses, le nombre de molécules de NaOH pour 2HCHO et 1 H_2O_2 (série 2-1-S).*

trations des réactifs. En effet, si l'on modifie les rapports des concentrations des réactifs, chaque réaction obéit encore à l'équation (1a), mais la valeur de A (comme celle de v_i qui lui est sensiblement parallèle) varie d'une façon complexe comme le montrent les figures 3, 4 et 5. Chacun de ces graphiques illustre l'effet de la variation de la concentration de l'un des réactifs, celles des deux autres étant constantes. La vitesse croît rapidement avec la con-

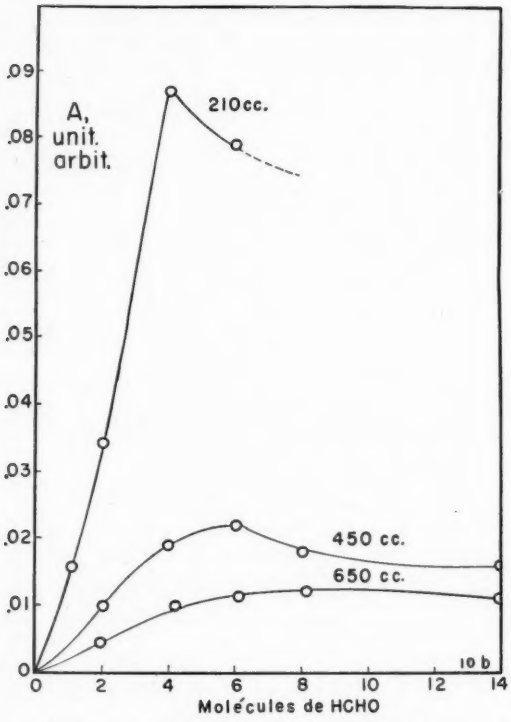


FIG. 4. *Valeurs de A en fonction de la proportion de formaldéhyde à 0°C. En abcisses, nombre de molécules de HCHO pour 1 H_2O_2 et 2 NaOH (série F-1-2). Les ordonnées sont à des échelles différentes.*

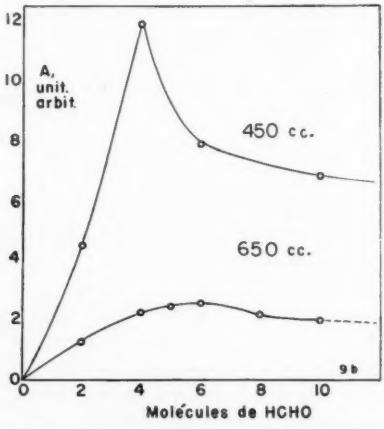


FIG. 4a. *Valeurs de A en fonction de la proportion de formaldéhyde à 25°C. En abcisses, nombre de molécules de HCHO pour 1 H_2O_2 et 2 NaOH (série F-1-2).*

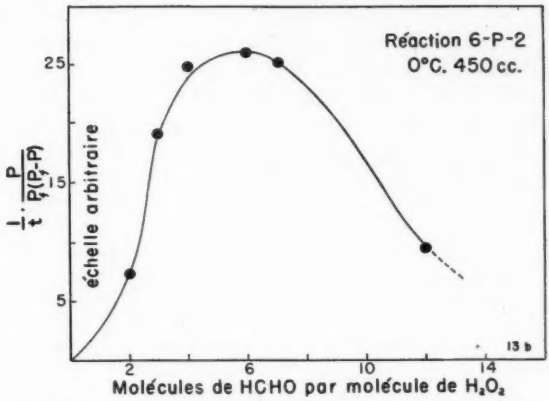


FIG. 5. *Influence de la proportion de l'eau oxygénée à 0°C., pour la série de compositions 6-P-2*

centration de la soude, mais une discontinuité apparaît à la composition $S/P = 4$ et $S/F = 2$. Lorsqu'on augmente la proportion de la formaldéhyde, on obtient une vitesse maximum à une composition qui dépend de la concentration de la solution. On observe aussi un maximum en fonction de la proportion de l'eau oxygénée. Les variations brusques d'allure semblent confirmer l'hypothèse de la formation d'un composé intermédiaire.

Le maximum de vitesse obtenu en faisant varier la proportion de la formaldéhyde se déplace en fonction de la concentration absolue de la façon indiquée sur les figures 4, 4*a* et 6. Il semble tendre vers la composition stoechiométrique 2-1-2 à concentration infinie.

Coefficient de température

Malgré ces complications, le coefficient de température est remarquablement simple. Si l'on porte log A contre l'inverse $1/T$ de la température absolue, on

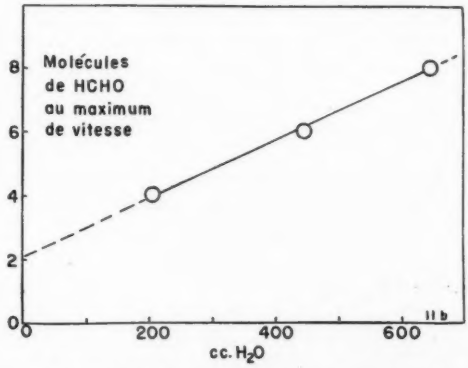


FIG. 6. *Déplacement de la position du maximum de vitesse en fonction de la dilution, pour la série de compositions F-1-2 à 0°C.*

obtient, pour des solutions de compositions très différentes, l'ensemble de droites parallèles de la figure 7. Le coefficient de température reste constant entre 0° et 25°C., est indépendant de la composition et correspond à une énergie d'activation $\Delta H_a = 14.5 \pm 0.3$ kcal./mole. A partir de cette valeur de ΔH_a et du temps de demi-réaction d'un mélange 2-1-2 à 0°C., nous avons calculé en supposant le second ordre, ce que seraient la valeur de k en litres $\times$ moles^{-1} $\times$ secondes^{-1} et celle du facteur S dans l'équation

$$k = Se^{-14500/RT}.$$

On obtient ainsi $S = 5 \times 10^{10}$, tandis que la valeur la plus fréquente est de l'ordre de 3×10^{11}. Cette réaction n'est donc pas affectée d'un facteur stérique

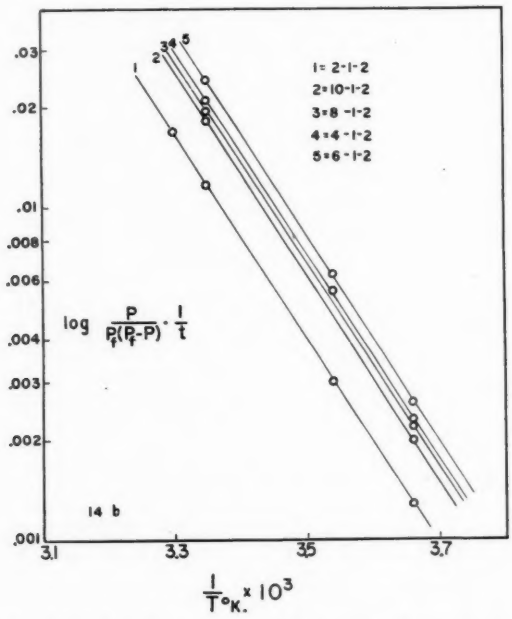


FIG. 7. *Variation de log A en fonction de 1/T °K. pour des réactions aux compositions 2-1-2, 10-1-2, 8-1-2, 4-1-2 et 6-1-2.*

appréciable et ne semble pas non plus comporter de longues chaînes. Naturellement, ceci n'indique que l'ordre de grandeur, puisque notre facteur A varie suivant la composition du mélange.

Décomposition du peroxyde de diméthylol

D'après Rieche et Meister (17, 18), le peroxyde de diméthylol (19) se décompose en solution alcaline suivant:

$$HOCH_2-O-O-CH_2OH + 2NaOH = 2HCOONa + 2H_2O + H_2.$$

Nous avons préparé ce peroxyde par la méthode de Fentón (6) et obtenu comme lui des cristaux fondant à 63-65°C. Des mesures cryoscopiques faites sur ce composé ont montré qu'il se trouve à l'état monomérique en solution aqueuse, les poids moléculaires trouvés étant de 87 et 98, tandis que celui de ce peroxyde est de 94.

Dissous dans l'eau et additionné de soude pour donner un mélange de composition équivalente à 2-1-2, ou de soude et de formaldéhyde pour donner 4-1-2, ce composé PF_2 nous a donné des réactions dont les courbes se superposent exactement à celles des mélanges 2-1-2 et 4-1-2 correspondants (Fig. 8).

Du point de vue de l'interprétation de notre réaction, ces résultats peuvent s'expliquer de deux façons: (*a*) en solution aqueuse, P et $2F$ forment PF_2 qui réagit ensuite avec la soude pour donner de l'hydrogène et les autres produits

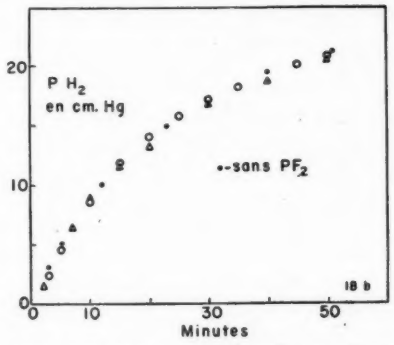


FIG. 8. *Identité des taux de deux réactions du peroxyde de diméthylol PF_2 additionné de soude, et d'un mélange de composition correspondante (2-1-2) à 0°C.*

ou bien, (*b*) PF_2 en solution alcaline se dissocie en $P + 2F$, qui réagissent avec la soude suivant un mécanisme inconnu. La seconde hypothèse nous ramène au cas trivial où PF_2 servirait seulement à fournir les ingrédients de la réaction.

Cryoscopie de mélanges d'eau oxygénée et de formaldéhyde

Dans l'espoir de résoudre le problème du complexe intermédiaire, posé par ces derniers résultats et par les discontinuités des courbes des figures 3, 4 et 4*a*, nous avons amorcé une étude cryoscopique des mélanges d'eau oxygénée et de formaldéhyde en solution aqueuse. Il a été malheureusement impossible de faire porter cette étude sur des solutions fortement alcalines parce que la réaction serait très rapide aux concentrations où il faut opérer pour observer des abaissements suffisants du point de congélation. Nous avons ajouté seulement la trace de soude nécessaire pour obtenir la dépolymérisation de la paraformaldéhyde.

Une solution de 0.33 mole/litre de formaldéhyde a donné un abaissement cryoscopique de 0.64°C. correspondant au poids moléculaire normal. La figure 9 montre ce qui arrive lorsqu'on ajoute à cette solution de l'eau oxygénée. A

mesure que la concentration de l'eau oxygénée augmente, l'abaissement cryoscopique décroît au lieu de croître, indiquant une diminution du nombre des molécules présentes dans la solution. A la proportion de 1 molécule d'eau oxygénée pour 3 de formaldéhyde, on atteint un abaissement cryoscopique minimum égal aux 3/4 de celui de la formaldéhyde seule. A première vue, on pense à la formation d'un complexe PF_3, mais ceci impliquerait que l'abaissement cryoscopique soit réduit à 1/3 de sa valeur initiale. Il s'agit plus vraisemblablement d'un équilibre entre les espèces PF, PF_2, PF_4, qui sont des composés connus.

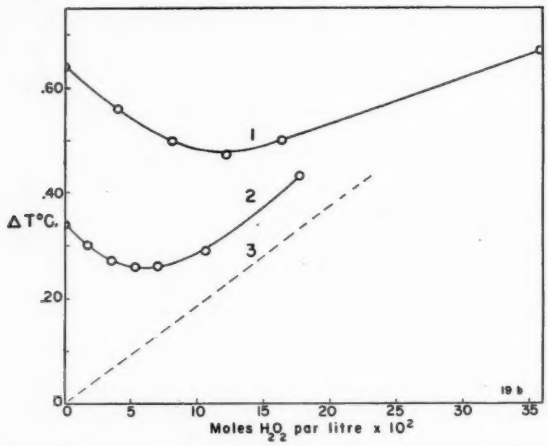


FIG. 9. *Variations de l'abaissement du point de congélation de deux solutions de formaldéhyde 0.33 molaire (1) et 0.165 molaire (2) en fonction de la concentration d'eau oxygénée ajoutée. La droite (3) indique l'abaissement que produirait l'eau oxygénée seule.*

Une étude détaillée de cet effet révélerait peut-être une correspondance significative entre ces minima cryoscopiques et les maxima des taux de réaction. Toutefois, l'apparition d'une légère opalescence dans la solution aux concentrations les plus élevées d'eau oxygénée laisse croire qu'il y avait formation d'un précipité colloïdal. En présence d'un tel effet, on ne peut attribuer une signification univoque à la position du minimum cryoscopique.

Discussion

Les résultats exposés ci-dessus révèlent que la réaction est très complexe. Si le coefficient de température est constant pour des réactions effectuées aux mêmes concentrations, par contre l'ordre par rapport aux concentrations initiales n'est pas celui que l'on obtient par rapport au temps. Il semble bien qu'on soit en présence de la décomposition bimoléculaire d'un intermédiaire dont la concentration ou la réactivité est une fonction complexe des concentrations relatives initiales.

A première vue, l'intermédiaire le plus vraisemblable est le peroxyde de dimé-

thylol, dont la formation (6, 17) et la décomposition (17) suivant l'équation:

$$2HCHO + H_2O_2 \rightarrow HOCH_2OOCH_2OH \xrightarrow{2NaOH} 2HCOONa + 2H_2O + H_2$$

sont connues. Nous avons montré que le taux de décomposition de ce peroxyde est égal au taux de réaction du mélange équivalent de formaldéhyde et d'eau oxygénée, mais les positions des minima cryoscopiques et la présence dans les solutions les plus concentrées d'un trouble suggérant la formation d'un polymère laissent entrevoir une situation beaucoup plus compliquée.

L'influence de la concentration de la soude est relativement simple. Le genou de la figure 3 correspond au point d'équivalence de la neutralisation de la formaldéhyde hydratée considérée comme acide faible:

$$HCHO \xrightarrow{H_2O} H_2C(OH)_2 \xrightarrow{2NaOH} H_2C(ONa)_2$$

Le sel de sodium, d'ailleurs fortement hydrolysé, est probablement la forme réactive de la formaldéhyde.

Puisque nous avons postulé l'intervention d'un intermédiaire du type du peroxyde de diméthylol, rappelons que ces peroxydes du type ROOR ont tendance à se trouver en solution sous la forme de radicaux libres RO· possédant un oxygène monovalent (4). En supposant que cette dissociation soit pratiquement complète dans les conditions de nos expériences, la recombinaison de deux de ces radicaux nous donnerait une réaction du second ordre. Suivant un mécanisme analogue à celui que Pearl (16) a proposé pour l'oxydation de la vanilline, ces étapes essentielles seraient les suivantes:

$$HCHO + H_2O \rightarrow H_2C\begin{matrix}OH\\OH\end{matrix} \xrightarrow{NaOH} H_2C\begin{matrix}O^-\\OH\end{matrix} + H_2O + Na^+$$

$$H_2C\begin{matrix}O^-\\OH\end{matrix} + Na^+ + HOOH \rightarrow H\dot{C}\begin{matrix}O^-\\OH\end{matrix} + OH^\cdot + H_2O + Na^+$$

$$2H\dot{C}\begin{matrix}ONa\\OH\end{matrix} \rightarrow 2\ HC\begin{matrix}ONa\\O\end{matrix} + H_2.$$

On suppose que les deux premières étapes sont rapides et complètes, laissant libre un excès éventuel soit de formaldéhyde, soit d'eau oxygénée contenant beaucoup d'ions OOH^-.

Ces substances en excès sont susceptibles d'inhiber la réaction principale en réagissant avec les radicaux $OH^\cdot$ et $H\dot{C}(OH)O^-$. On peut supposer, par exemple, que ce dernier radical sert à amorcer et à propager la formation d'un polymère d'addition

$$H\dot{C}(OH)O^- + n\ HOCH_2OH \rightarrow n\ H_2O + HOCH_2(OCH_2)_{n-1}\dot{C}H(OH)O^-$$

dans lequel la formaldéhyde se trouverait protégée contre la réaction principale. Rieche et Meister (18) ont démontré l'existence de tels polymères et leur présence rendrait compte du léger trouble observé dans nos solutions. Rappelons

que l'hydrolyse par la soude des polymères de la formaldéhyde est affectée d'une énergie d'activation de l'ordre de 15 kcal./mole (13) voisine de celle que nous avons mesurée.

Il se peut que les maxima de vitesse résultent de l'action de deux processus antagonistes: l'un de polymérisation et l'autre de dépolymérisation par la soude. Ce dernier libérerait les espèces réactives et imposerait peut-être son énergie d'activation. Cependant, il semble difficile d'expliquer les discontinuités des courbes et l'ordre de la réaction si l'on n'assigne pas le rôle central à un complexe intermédiaire relativement simple.

Rappelons en terminant que les réactions du permanganate de potassium avec l'eau oxygénée (2) et l'acide oxalique (5) présentent aussi des vitesses maxima difficiles à expliquer.

Remerciements

L'un de nous (J. B. J.) exprime ses plus vifs remerciements à la Consolidated Mining and Smelting Company, pour la bourse de recherches dont il a bénéficié.

References

1. Bach, A. et Generosov, A. Ber. 55: 3560. 1922.
2. Bailey, K. C. et Taylor, J. T. J. Chem. Soc. 994. 1937.
3. Blank, O. et Finkenbeiner, H. Ber. 31: 2979. 1898.
4. Branch, G. E. K. et Calvin, N. M. The theory of organic chemistry. Prentice-Hall, New York. 1941. p. 332.
5. Chang, T. L. et Riesenfeld, E. H. Z. anorg. u. allgem. Chem. 230: 239. 1937.
6. Fenton, H. T. H. Proc. Roy. Soc. (London), A, 90: 492. 1914.
7. Fry, H. S. et Payne, J. H. J. Am. Chem. Soc. 53: 1973, 1975. 1931.
8. Geisow, H. Ber. 37: 515. 1904.
9. Glasstone, S. et Hickling, A. Electrolytic oxidation and reduction. Chapman and Hall, London, 1935. Chem. Revs. 25: 407. 1939.
10. Harden, A. Proc. Chem. Soc. 15: 15B. 1899.
11. Hatcher, W. H. et Holden, G. W. Trans. Roy. Soc. Can. 20: 385. 1926.
12. Kastle, J. H. et Loevenhart, A. S. J. Am. Chem. Soc. 21: 262. 1899.
13. Loebering, J. et Rank, V. Ber. 70: 2331. 1937.
14. Lyford, C. A. J. Am. Chem. Soc. 29: 1227. 1907.
15. Mueller, E. et Tagekani, S. Z. Elektrochem. 34: 904. 1928.
16. Pearl, I. A. J. Am. Chem. Soc. 67: 1628. 1945; 68: 429. 1946.
17. Rieche, A. et Meister, R. Ber. 66: 718. 1933.
18. Rieche, A. et Meister, R. Ber. 68: 1465. 1935.
19. Traube, M. Ber. 16: 123. 1883.
20. Wieland, H. et Wingler, A. Ann. 431: 301. 1923.
21. Wieland, H. et Richter, D. Ann. 486: 226. 1931; 495: 284. 1932.
22. Wirtz, K. et Bonhoeffer, N. F. Z. physik. Chem. B, 32: 108. 1935.

THE OXIDATION OF CARBON MONOXIDE BY SOLID SILVER PERMANGANATE REAGENTS

IV. CHEMICAL COMPOSITION IN RELATION TO ACTIVITY[1]

By Morris Katz, L. G. Wilson, and R. Riberdy

Abstract

The activity after progressive aging and thermal treatment of silver permanganate supported on zinc oxide and beryllium oxide was found to be closely related to the extent of decomposition of the crystalline silver permanganate. The silver salt decomposed, with loss of active oxygen, to a relatively insoluble, amorphous product resembling a manganito-manganate of the composition, $Ag_2O \cdot Mn_2O_5$ or $[AgMnO_3]_2$. From a mass balance of solid–gas reactants in a reacting column, it was demonstrated that in the most active stage, during which the efficiency of oxidation did not fall below 90%, the course of the reaction was about evenly divided between stoichiometric and catalytic processes.

Introduction

Investigations on the heterogeneous oxidation of carbon monoxide over silver permanganate supported on various metallic oxides, in particular zinc oxide, have been reported by Katz *et al.* (6, 7). The methods of preparation in relation to activity of the granules have been described elsewhere (4). For each metallic oxide support there is apparently an optimum mole composition range of the silver salt and oxide for maximum life and activity.

It was found by Katz and Halpern (7) that some silver permanganate reagents (notably those containing nickel oxide and cobalt oxide, Co_2O_3) were highly reactive towards carbon monoxide when freshly prepared, but this activity declined rapidly after several days' thermal treatment at 60°C. In other instances there was a more or less steady decline with prolonged heating. However, some preparations, which were only moderately active initially, showed an improvement after several days' thermal treatment, for example—a lead peroxide, Pb_3O_4, preparation. Improved thermal characteristics were found to depend not only upon the type of metallic oxide support, but also on the mole composition of oxide and silver salt.

Dry, crystalline silver permanganate slowly undergoes a change at ordinary temperatures, with spontaneous evolution of oxygen, gradually losing its water solubility. The reaction is autocatalytic in nature and is characterized by an induction period. The decomposition velocity increases with rising temperature and the rate becomes appreciable above 90° to 100°C. The reaction is catalyzed by small amounts of water and by the addition of end products, as well as by some metallic oxides (5, 10, 11).

Various end products have been reported in the literature. Gorgeu found that moist silver permanganate decomposed thermally to a product,

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$AgMnO_3.\frac{1}{2}H_2O$ (2). Sieverts and Theberath (11) confirmed substantially the findings of Gorgeu, although they found that the inactive oxygen in the decomposition product was always somewhat lower than that required by the above formula. In confirmation of this, the moist silver permanganate yielded slightly more oxygen than that required according to the simple equation, $AgMnO_4 \rightarrow AgMnO_3 + \frac{1}{2}O_2$.

Hein (5) found that dry thermal decomposition of the silver permanganate at 100°C. yielded an amorphous water-insoluble black product, $[8Ag_2O \cdot 15MnO_2 \cdot MnO_3 \cdot O]$. When the moist salt was decomposed at 100°, the end product was also a graphite-like, water-insoluble material corresponding to the composition $[8Ag_2O \cdot 15MnO_2 \cdot MnO_3 \cdot 6O \cdot 8H_2O]$, or $AgMnO_{2.94} \cdot \frac{1}{2}H_2O$. The water is apparently firmly bound to the molecule as it is not removed by drying over concentrated sulphuric acid.

In the present paper the results of a study of the intermediate and end products after prolonged aging, thermal and carbon monoxide treatment are presented in an attempt to elucidate the course of the reaction between the silver salt and carbon monoxide. The solid products were investigated by chemical analysis and X-ray diffraction.

Materials and Analytical Methods

The reagents employed in these experiments contained silver permanganate and zinc oxide or beryllium oxide in definite molecular proportions. The method of preparation has been outlined in previous publications (4, 7). Briefly, crystals of silver permanganate are deposited on the oxide carrier, *in situ*, by the action of a solution of potassium permanganate on concentrated silver nitrate solution intimately mixed with the oxide to form a paste. In contrast to these reagents, samples were also prepared by mechanical mixing of pure silver permanganate and zinc oxide, in the presence of a small amount of water, the mixture pressed at 10 tons per sq. in., and the pressed cake cured and broken up into granules, as in the regular method of preparation. These samples have been denoted as "mechanical mixture" in the tables. All samples were dried at room temperature and screened from -8 to $+24$ mesh Tyler.

The apparatus and experimental methods employed in the carbon monoxide activity tests, including the preparation and purification of the carbon monoxide, measurement of the initial and escaping concentrations of gas in the reaction cell and other details, have already been described in previous publications (8, 3, 7). The glass reaction cell was 2.84 cm. in diameter and was usually filled with granules to a column length of 4 cm. The gas and air mixtures were passed through this bed at a space velocity of 625 cm. $min.^{-1}$, at concentrations of 1.0% or 0.5%, as indicated. The breakdown time of the bed was chosen arbitrarily as the time at which the escaping concentration exceeded a value which was 10% of the influent concentration.

Standard methods of analysis were employed in determining the chemical

composition of the various preparations before and after treatment (1). Total silver was determined gravimetrically as silver chloride (1, pp. 821-23). Total manganese was estimated by the Volhard method or gravimetrically as the pyrophosphate (1, pp. 560-61). Zinc oxide was separated as zinc sulphide and titrated in acid solution against potassium ferrocyanide (1, pp. 1060-61). Beryllium was separated from the silver, precipitated as the hydroxide, redissolved in 10% sodium bicarbonate to remove manganese hydroxide, and reprecipitated as the hydroxide. It was then determined gravimetrically as the oxide after ignition (1, pp. 140-41).

Crystalline silver permanganate was determined by treatment of the sample with distilled water, filtering through a Gooch crucible packed with purified asbestos wool, and analysis of the filtrate by the volumetric method, using an excess of standard sodium oxalate solution in the presence of sulphuric acid, the excess being titrated with standard potassium permanganate solution.

Higher manganese oxides, containing "active" oxygen, were determined by treating a sample directly with an excess of standard sodium oxalate solution and 5% sulphuric acid, agitating the mixture heated to 60 to 65°C. until the sample dissolved and then back titrating the excess oxalate with 0.1 *N* potassium permanganate solution.

The portion of the total silver and manganese not accounted for in the water-soluble fraction was expressed, empirically, as $AgMnO_3$ or $Ag_2O \cdot Mn_2O_5$.

X-Ray Diffraction Method

A standard X-ray diffraction technique was employed to study crystallinity. Spectra were photographed for stationary and for rotating specimens in flat and cylindrical cameras respectively. From these spectra, the substances present, their crystalline sizes, and the relative proportions of the various compounds, could be noted. A copper target diffraction tube was used in all cases.

Results

Effect of Aging on Activity

The silver permanganate reagents undergo a change during progressive aging. The magnitude of this trend is indicated by the activity data given in Table I. The standard preparation of $AgMnO_4$-ZnO, 31-69 mole %, showed little, if any, loss in activity towards carbon monoxide after six months, and only a small decrease after 14 months' aging at room temperature. The activity of all preparations in Table I decreased markedly on prolonged heating at 60°C. The standard zinc oxide supported reagents were markedly superior to those prepared by mechanical mixing of the ingredients. The beryllium oxide preparations, although possessing good activity initially, were found to be inferior to those containing zinc oxide when subjected to aging or heat treatment.

Influence of Aging on Chemical Composition

The influence of aging and thermal treatment on chemical composition of a series of standard preparations containing zinc oxide or beryllium oxide is

TABLE I

EFFECT OF THERMAL TREATMENT ON ACTIVITY

Reagent	Composition, mole % $AgMnO_4$	Age at room temp., months	Thermal treatment at 60°C., hr.	Activity towards carbon monoxide, space velocity, 625 cm. $min.^{-1}$, column length, 4.0 cm.	
				Concentration, %	Breakdown time to 90% efficiency, min.
$AgMnO_4$–ZnO standard preparation	31	0.75		1.0	96
		6		1.0	93
		8		1.0	86
		8	160	1.0	24
		14		0.50	143
		0.75	506	0.50	81
$AgMnO_4$–ZnO standard preparation	33	0.5		1.0	94
	36	0.75		1.0	103
		2	160	1.0	51
		0.75	506	0.50	65
$AgMnO_4$–ZnO "mechanical mixture"	31	0.5		1.0	63
		0.5	160	1.0	Nil
		0.75		1.0	73
		2	148	1.0	29
$AgMnO_4$–BeO standard preparation	15	0.5		0.50	108*
		5	160	0.50	Nil
$AgMnO_4$–BeO standard preparation	25	0.5		0.50	106*
		6		0.50	86

* *Breakdown time for a 3.0 cm. column length.*

illustrated in Table II. The content of the water-soluble, crystalline, silver permanganate fraction decreases with time or thermal treatment as conversion takes place to a relatively insoluble amorphous reaction product. This product

TABLE II

CHEMICAL COMPOSITION DURING THERMAL AGING OF STANDARD PREPARATIONS

Description of sample	Age at room temp., months	Hours of heating at 60 °C.	Composition in % by weight			
			$AgMnO_4$	$AgMnO_3$	ZnO	Total
$AgMnO_4$–ZnO, 36-64 mole %	2	Nil	56.07	3.16	40.80	100.03
"	2	160	25.98	32.60	41.40	99.98
"	4*	160	14.72	42.23	41.92	98.87
$AgMnO_4$–ZnO, 31-69 mole %	9	Nil	46.40	6.20	46.68	99.28
"	14	Nil	44.04	8.31	47.15	99.50
					BeO	
$AgMnO_4$–BeO, 25-75 mole %	6	Nil	58.12	15.06	28.18	101.36
$AgMnO_4$–BeO, 15-85 mole %	5	160	0.00	53.52	46.87	100.39

* *Sample aged two months longer at room temperature after thermal treatment at 60°C.*

PLATE I

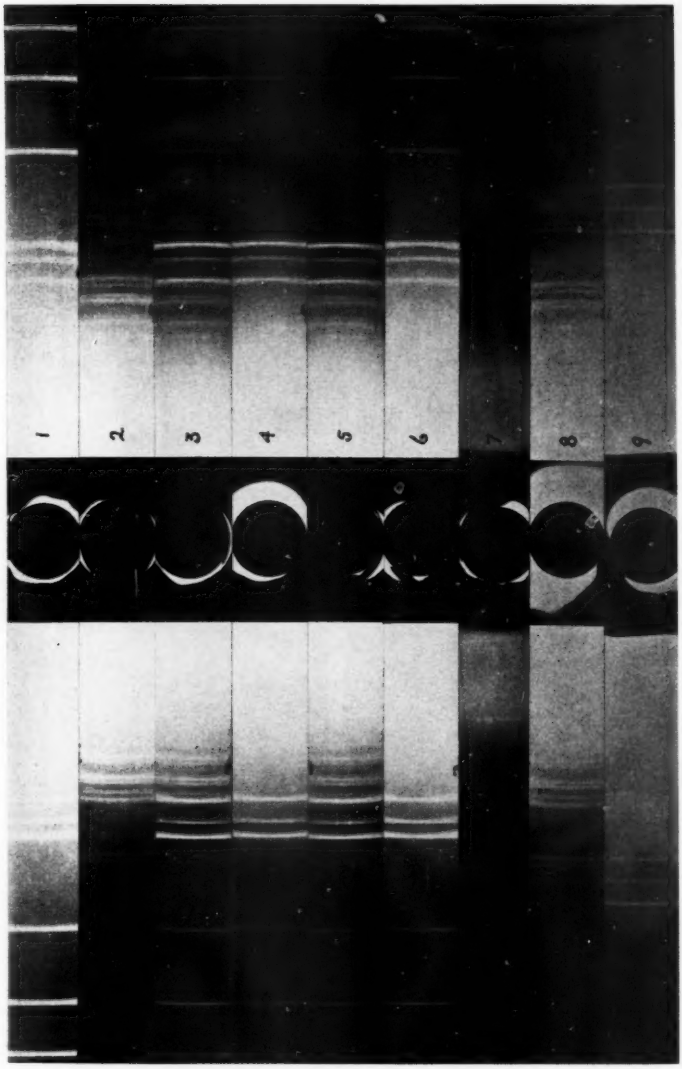
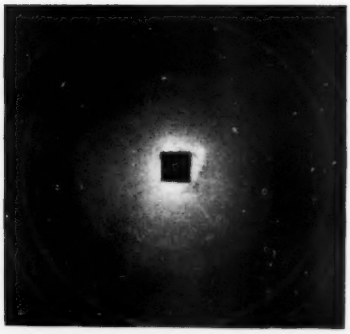
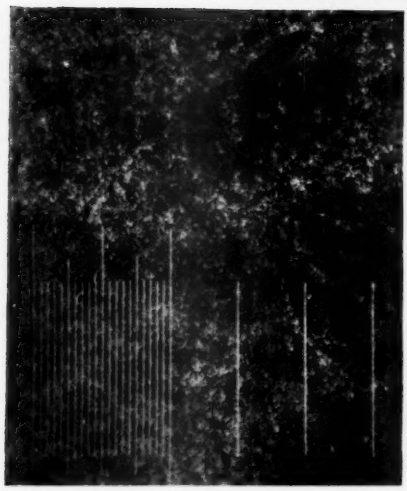


FIG. 1. *Pure zinc oxide spectrum.*
FIG. 2. *Pure silver permanganate spectrum.*
FIG. 3. *Reagent containing originally 31 mole % $AgMnO_4$–69 mole % ZnO, nine months old.*
FIG. 4. *A mechanical mixture containing originally 31 mole % $AgMnO_4$–69 mole % ZnO, heated for 148 hr. at 60°C.*
FIG. 5. *A mechanical mixture containing originally 31 mole % $AgMnO_4$–69 mole % ZnO, three weeks old.*
FIG. 6. *Same reagent as in Fig. 3, but X-ray pattern taken after sample had been reacted completely with carbon monoxide as in Experiment No. 3.*
FIG. 7. *Pure beryllium oxide spectrum.*
FIG. 8. *Beryllium oxide reagent, containing originally 15 mole % $AgMnO_4$, not heat treated.*
FIG. 9. *Beryllium oxide reagent, same original composition as in Fig. 8, aged 160 hr. at 60°C.*

PLATE II





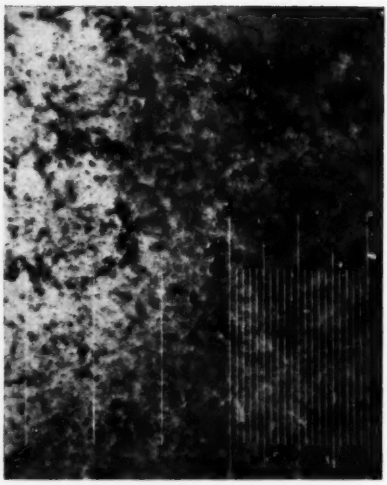


FIG. 10. *Zinc oxide reagent, containing originally 31 mole % $AgMnO_4$, aged eight months at room temperature, and then aged 160 hr. at 60°C. "Spotty" rings are from $AgMnO_4$.* FIG. 11. *Photomicrograph of standard zinc oxide – silver permanganate reagent, showing particle size. (Small scale division = 10 μ.)* FIG. 12. *Photomicrograph of a mechanical mixture of zinc oxide and 30% silver permanganate. (Small scale division = 10 μ.)*

contains available oxygen and from the analytical results is best represented as a manganito-manganate of the composition $Ag_2O \cdot Mn_2O_5$ or $AgMnO_3$, closely resembling the product obtained by Hein in the thermal decomposition of the moist salt at 100°C., i.e., $AgMnO_{2.94} \cdot \frac{1}{2}H_2O$. Compounds of this type containing available oxygen to manganese in the atomic proportions of 1.5 to 1.6 have been discussed by Mellor (9).

It is also evident from Table II that the reagent containing zinc oxide as the carrier is much more stable than the one containing beryllium oxide. Prolonged thermal treatment at 60°C. of the beryllium oxide reagent produced complete conversion of the silver permanganate fraction to lower oxidation products, whereas corresponding treatment of the zinc oxide reagent resulted in about 54% decomposition of the permanganate.

TABLE III

CHEMICAL COMPOSITION DURING AGING OF A MECHANICAL MIXTURE

Description of sample	Age at room temp., months	Hours of heating at 60°C.	Composition in % by weight			
			$AgMnO_4$	$AgMnO_3$	ZnO	Total
$AgMnO_4$–ZnO, 31-69 mole %	0.75	Nil	50.00	5.53	44.40	99.93
"	4	Nil	20.30	32.07	45.39	97.76
"	6	Nil	10.21	43.45	45.73	99.39
"	6	148	0.40	53.0	46.40	99.80
Standard preparation, $AgMnO_4$–ZnO, 31-69 mole %	9	Nil	46.40	6.20	46.68	99.28

This retarding influence of zinc oxide on the thermal decomposition of the silver salt is not achieved by mere mechanical mixing and pressing of the ingredients of the reagent, as shown in Table III. The aging of a mechanical mixture is poor by comparison with that of a standard preparation, as judged by the relatively rapid disappearance of the crystalline silver salt fraction.

The data in Tables I to III, inclusive, indicate that there is a direct relation between the extent of decrease of the silver permanganate concentration and the reduction in breakdown time or oxidation activity towards carbon monoxide on prolonged aging. Samples in which the soluble silver salt had been decomposed completely showed virtually no activity.

The X-ray diffraction patterns of a number of samples are given in the photographs, Figs. 1-10 inclusive. Pure zinc oxide and pure silver permanganate are shown in Figs. 1 and 2 respectively. Fig. 7 indicates the spectrum of pure beryllium oxide. The remaining photographs show the spectra of various samples after treatment. Reagents which exhibited a high reactivity with carbon monoxide showed also that a large proportion of the original silver permanganate was still present in the crystalline state. Those samples which had lost their oxidizing power for carbon monoxide showed only the spectrum of the metallic oxide carrier. The products formed from the breakdown of the permanganate are apparently a chemically bound mixture of amorphous oxides

TABLE IV

MASS BALANCE IN HETEROGENEOUS REACTION SYSTEM

	Run No. 1	Run No. 2	Run No. 3
Weight of sample, gm.	62.0	65.0	101.0
Weight of $AgMnO_4$ in sample before reaction, gm.	27.30	35.20	46.86
Weight of $AgMnO_4$ in sample after reaction, gm.	12.94	9.89	0.06
Weight of CO oxidized, gm.	4.131	4.754	9.05
Weight of O_2 required to oxidize CO, gm.	2.361	2.717	5.17
Weight of O_2 lost by sample, gm.	1.100	1.495	3.587
Weight of $AgMnO_4$ which reacted with CO, gm.	14.36	25.31	46.80
Weight of O_2 supplied by $AgMnO_4$, gm., according to the reaction:			
(*A*) $2\ AgMnO_4 \longrightarrow 1.5\ O_2(+Ag_2O + 2\ MnO_2)$	1.520	2.678	4.952
(*B*) $2\ AgMnO_4 \longrightarrow O_2(+2AgMnO_3)$	1.013	1.785	3.302

which from chemical analytical evidence may be represented as a silver manganito-manganate. The products obtained by Hein from the thermal decomposition of both the dry and the moist salt at elevated temperatures were also amorphous (5).

The difference in particle size between a standard reagent containing silver permanganate deposited on the zinc oxide by chemical reaction, *in situ*, and that of a mechanical mixture is illustrated by the photomicrographs in Figs. 11 and 12. The standard reagent shows a much smaller particle size and more uniform distribution.

Nature of Heterogeneous Oxidation of Carbon Monoxide

A mass balance was made of the reactants during the oxidation of carbon monoxide in a reacting column containing various standard samples of granules. The mass balance in three experiments is shown in Table IV. The amount of oxygen removed from the silver permanganate during the oxidation represents the stoichiometric reaction between carbon monoxide and the solid reagent. Any additional oxidation would be catalytic, at the expense of oxygen

TABLE V

STOICHIOMETRIC AND CATALYTIC OXIDATION OF CARBON MONOXIDE

	Run No. 1		Run No. 2		Run No. 3	
	% Oxidation					
	Stoichiometric	Catalytic	Stoichiometric	Catalytic	Stoichiometric	Catalytic
1. Calculated on the actual weight loss of oxygen	46.63	53.37	55.02	44.98	69.4	30.6
2. Calculated on the amount of oxygen supplied by $AgMnO_4$, according to the reaction:						
(*a*) $2\ AgMnO_4 \longrightarrow 1.5\ O_2$ $(+Ag_2O + 2MnO_2)$	64.36	35.64	98.57	1.43	95.77	4.23
(*b*) $2AgMnO_4 \longrightarrow O_2$ $(+2AgMnO_3)$	42.90	57.10	65.70	34.30	63.85	36.15

in the gas phase. Two alternative reaction paths for liberation of oxygen from the silver salt are shown in Table IV. Reaction (*B*) is more nearly in accord with the actual weight of oxygen released from each sample during reaction with carbon monoxide and is also more nearly representative of the solid products formed during aging and thermal decomposition, as shown by chemical analysis.

The calculated percentage conversion of CO to CO_2 according to stoichiometric and catalytic reactions is given in Table V. The chemical composition of each sample immediately before and after treatment with carbon monoxide

TABLE VI

COMPOSITION AFTER REACTION WITH CARBON MONOXIDE

Original composition of sample in % by weight	Age of sample	Quantity of carbon monoxide oxidized, gm.	Composition in % by weight			
			$AgMnO_4$	$AgMnO_3$	ZnO	Total
Run No. 1						
$AgMnO_4$–ZnO, 55.6-44.4	14 mos.	Nil	44.04	8.31	47.15	99.50
"	14 mos.	4.13	20.88	31.11	47.86	99.85
Run No.2						
$AgMnO_4$–ZnO, 57.8-42.2	12 days	Nil	54.15	1.00	44.87	100.02
"	12 days	4.75	15.21	35.85	45.84	99.60
Run No. 3						
$AgMnO_4$–ZnO, 55.6-44.4	9 mos.	Nil	46.40	6.20	46.68	99.28
"	9 mos.	9.05	0.06	51.23	47.86	99.15

is shown in Table VI. The stoichiometric reaction may be represented best by the following equation

$$2\ AgMnO_4 + 2\ CO \longrightarrow 2\ CO_2 + Ag_2O \cdot Mn_2O_5. \quad (1)$$

An alternative possibility is

$$16\ AgMnO_4 + 17\ CO + 8\ H_2O_{vapor} \longrightarrow 17\ CO_2 + [8\ Ag_2O \cdot 15\ MnO_2 \cdot MnO_3 \cdot 3O_2 \cdot 8\ H_2O]. \quad (2)$$

The solid reaction product in the second equation represents the thermal decomposition product reported by Hein (5). It has been mentioned that a little water vapor is necessary to initiate the oxidation of carbon monoxide, although the standard reagent is substantially nonhygroscopic. Samples in equilibrium with an atmosphere at 89% relative humidity for 48 hr. at room temperature showed a moisture absorption of only 0.09% by weight, whereas the thermally decomposed product absorbed 0.35 to 0.46% water.

From the data in Tables IV and V, it is evident that when the efficiency of oxidation in a reacting column does not go below 90%, the percentage conversion of carbon monoxide to carbon dioxide is, roughly, evenly divided between the stoichiometric and catalytic processes. Below 90% efficiency, a greater proportion of the carbon monoxide is oxidized stoichiometrically as the active sites in the granular mass become depleted or "poisoned" with time.

Because of this direct reaction with the silver salt, the granular reagent oxidizes carbon monoxide effectively in gas mixtures which do not contain gaseous oxygen. When a stream of 1.0% carbon monoxide in nitrogen containing about 0.1% oxygen was passed through a column of granules of standard reagent consisting of 31 mole % silver permanganate on zinc oxide, under the test conditions indicated in Table I, the breakdown time was 77 min. When the oxygen in the nitrogen was removed completely, by passage through a purification train, the breakdown time was 68.5 min.

Discussion

There is a close correlation between the disappearance of the crystalline state of the silver salt and loss of activity as shown by chemical analysis and X-ray diffraction. Decay in activity produced either by reaction with carbon monoxide or by thermal treatment is paralleled by the conversion of the crystalline silver salt to an amorphous reaction product of the approximate composition, $Ag_2O \cdot Mn_2O_5$.

Prout and Tompkins (10) have recently proposed that pure dry silver permanganate may decompose thermally, as follows, from kinetic data,

$$2\,AgMnO_4 \longrightarrow 1.5\,O_2 + Ag_2O \cdot 2\,MnO_2. \qquad (3)$$

Such a reaction, if applied to the stoichiometric oxidation of carbon monoxide, would imply the reduction of two molecules of the silver salt by three molecules of carbon monoxide. However, the above authors did not analyze the solid products of the thermal decomposition. Apart from this, the present work deals with a reaction which takes place in the presence of water vapor, consequently, only data dealing with the thermal decomposition or reduction of the moist salt are relevant.

The mechanism of the heterogeneous oxidation of carbon monoxide by these solid reagents is complex and there is, as yet, insufficient experimental evidence for a complete explanation. The stoichiometric reaction between carbon monoxide and the silver salt involves the reduction of one molecule of silver permanganate per molecule of carbon monoxide, based on the chemical analysis of the solid products. Some confirmation of this has been obtained from kinetic data (8). It is also supported by the data on thermal decomposition reported here.

References

1. FURMAN, N. H. Scott's standard methods of chemical analysis. Vol. 1. D. Van Nostrand Co. Inc., New York. 1939.
2. GORGEU, A. Bull. Soc. Chim. III, 7: 261. 1892.
3. GRANT, G. A., KATZ, MORRIS, and HAINES, R. L. Can. J. Technol. 29: 43. 1951.
4. GRANT, G. A., KATZ, MORRIS, and RIBERDY, R. Can. J. Technol. 29: 511. 1951.
5. HEIN, FR. Z. anorg. u. allgem. Chem. 235: 25. 1937.
6. KATZ, MORRIS, GRANT, G. A., and RIBERDY, R. Can. J. Research, B, 28: 799. 1950.
7. KATZ, MORRIS and HALPERN, SOPHIE. Ind. Eng. Chem. 42: 345. 1950.
8. KATZ, MORRIS and KATZMAN, J. Can. J. Research, F, 26: 318. 1948.
9. MELLOR, J. W. Inorganic and theoretical chemistry. Vol. 12. Longmans, Green & Co., New York. 1947. p. 284.
10. PROUT, E. G. and TOMPKINS, F. C. Trans. Faraday Soc. 42: 468. 1946.
11. SIEVERTS, A. and THEBERATH, H. Z. physik. Chem. 100: 463. 1922.

DIPHENYLCYANAMIDE DERIVATIVES[1]

By J. R. Robinson[2] and W. H. Brown

Abstract

Diphenylcyanamide is conveniently prepared by the action of cyanogen chloride on diphenylamine. When it is hydrolyzed in the presence of hydroxylated solvents, of the type ROH, stable isoureas are formed. These are readily hydrolyzed in aqueous alkaline medium to the corresponding urea derivative. Chlorination of diphenylcyanamide occurs in the *para* positions yielding a nitrogen analogue of DDT. This chlorinated derivative is only 1/80th as toxic as DDT to *Sitophilus granarius* (L).

Diphenylcyanamide (I) has been reported by von Braun (2), Werner (26), and Kurzer (10). Since only a few direct derivatives have been recorded (3, 4, 5, 9, 26), an investigation of this compound is of interest. For example, *p*, *p'*-dichlorodiphenylcyanamide (II) is a nitrogen analogue of 1,1-*bis*-(*p*-chlorophenyl)-2,2,2-trichlorethane (DDT) and might show insecticidal activity.

It has been found that diphenylcyanamide (I) is formed in excellent yield by treating diphenylamine (IX) with cyanogen chloride at 60°C. and 5 atm. pressure. When it is hydrolyzed in aqueous alkaline medium, N, N-diphenylurea (III) results. Michler (16) and Kurzer (11) have reported other methods for preparing this latter compound. When a solution of diphenylcyanamide (I) in methanolic potassium hydroxide is refluxed for 72 hr., diphenylamine (IX) is produced. If, however, the reaction is interrupted after two hours, N, N-diphenylmethylisourea (IV) is the major product. The isourea is readily converted to N, N-diphenylurea (III) by boiling with aqueous potassium hydroxide for 2.5 hr.

When ethanol is used as solvent instead of methanol, the corresponding ethylisourea (V) is readily obtained.

Pyrolysis of N, N-diphenylmethylisourea (IV) leads to the formation of N, N-diphenylurea (III) in poor yield.

Treatment of a chloroform solution of diphenylcyanamide (I) with chlorine gives *p*, *p'*-dichlorodiphenylcyanamide (II). This latter compound when refluxed in a solution of potassium hydroxide in methanol for two hours forms N, N-di-*p*-chlorophenylmethylisourea (VIII). If the reaction is allowed to proceed for 24 hr., *p*, *p'*-dichlorodiphenylamine (VII) results. An authentic sample of *p*, *p'*-dichlorodiphenylamine was prepared by the methods of Claus and Shaare (8) and Chapman (7).

Aqueous alkaline hydrolysis of *p*, *p'*-dichlorodiphenylcyanamide (II) gives N, N-di-*p*-chlorophenylurea (VI), which hydrolyzes with difficulty in dilute hydrochloric acid solution to *p*, *p'*-dichlorodiphenylamine (VII).

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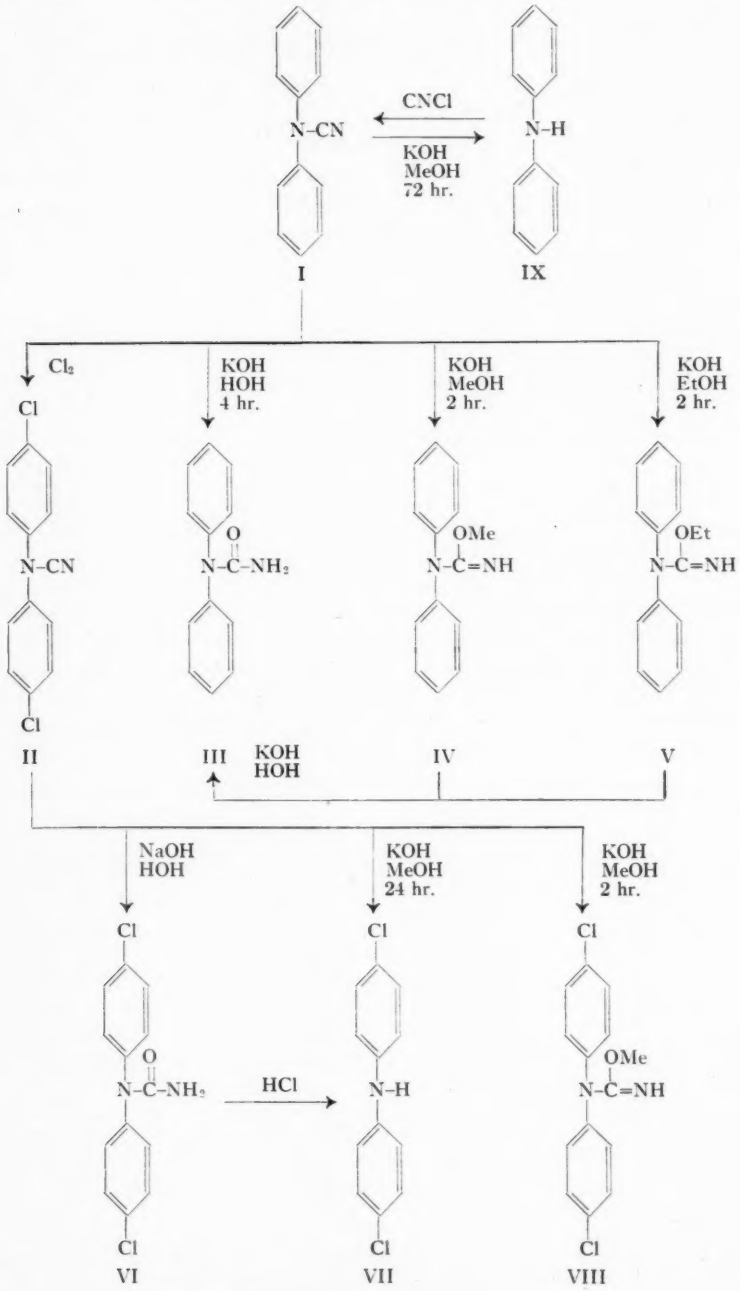
CNCl
KOH
MeOH
72 hr.
N–CN
N–H
I
IX
Cl₂
KOH
HOH
4 hr.
KOH
MeOH
2 hr.
KOH
EtOH
2 hr.
Cl
N–CN
Cl
O
N–C–NH₂
OMe
N–C=NH
OEt
N–C=NH
II
III
KOH
HOH
IV
V
NaOH
HOH
KOH
MeOH
24 hr.
KOH
MeOH
2 hr.
Cl
O
N–C–NH₂
HCl
N–H
OMe
N–C=NH
Cl
VI
VII
VIII

Discussion

Pinner (18) was among the first to report the synthesis of iminoethers.* Many investigators (1, 12, 13, 14, 15, 17, 19, 20, 21, 22, 23, 24, 25) have extended the field, but none of the N, N-diphenyl type is reported. McKee (13) predicted that disubstituted cyanamides would most readily add alcohols when catalyzed by the corresponding alkoxide ion, rather than by protons. The authors' findings with respect to diphenylcyanamide bear this out. Methanol adds to diphenylcyanamide in two hours in the presence of methoxide ion in 88% yield, whereas the reaction requires 80 days in the presence of hydrogen chloride to give a yield of 3.5%.

Toxicology

Tests† show that *p, p'*-dichlorodiphenylcyanamide (II) possesses an estimated median lethal concentration of about eighty times that of DDT to *Sitophilus granarius* (L). The other derivatives of diphenylcyanamide were also screened for activity, but did not indicate toxicity great enough to warrant quantitative investigation.

Browning *et al.* (6) have tested a number of N-substituted *p, p'*-dichlorodiphenylamines for biological activity. Their results show that groups such as methyl, trichloromethyl, and trichloroacetyl do not increase the toxicity of the parent compound. The cyanide group does have an enhancing effect according to the present authors' findings.

Experimental‡

Diphenylcyanamide

After treating 34 gm. (0.2 mole) of diphenylamine with 12.3 gm. (0.2 mole) of cyanogen chloride at 60°C. for one and one-half hours in a capped ginger ale bottle, the reaction mixture was allowed to stand at room temperature for 24 hr. As a safety precaution, the bottle was kept immersed in a steel-jacketed water bath throughout the course of the reaction.

The ether-soluble portion of the resulting solid was crystallized three times from absolute ethanol, giving 17.5 gm. (90%) of a white granular solid, which melted at 73°C.

p, p'-Dichlorodiphenylcyanamide

Chlorine was bubbled into a solution of 7.76 gm. (0.04 mole) of diphenylcyanamide in 30 ml. of chloroform at 5-6°C. for one hour. The solution was

* *These compounds are recorded in the literature under various names; imino- or imidoethers imino- or imidoesters and esters of imidic acids. Those of the type* $R_2N\text{-}C(OR') = NH$ *are called isocarbamides, carbimidoethers (esters), pseudoureas, or isoureas.*

† *Material tested by the Entomology Section, Defence Research Board Experimental Station, Suffield, Alta.*

‡ *All melting points have been corrected against reliable standards. Combustion analyses are by A. E. Ledingham, Dominion Rubber Research Laboratories, Guelph, Ont.; R. G. Pyke, Department of Chemistry, University of Toronto, and J. F. Eagen, National Research Council, Ottawa.*

treated with 10% aqueous sodium carbonate, washed with water, and finally dried over anhydrous sodium sulphate. When the chloroform was distilled off, there remained 12.1 gm. of golden yellow oil which crystallized spontaneously. Crystallization from ethanol yielded 6.1 gm. (58%) of long colorless needles melting at 114-115°C. Calc. for $C_{13}H_8Cl_2N_2$: C, 59.31; H, 3.06; Cl, 26.94; N, 10.65%. Found: C, 59.06; H, 3.06; Cl, 26.90; N, 10.64%.

N, N-Diphenylurea

To a solution of 1.12 gm. (0.02 mole) of potassium hydroxide in 100 ml. of water was added 3.88 gm. (0.02 mole) of diphenylcyanamide, and the mixture refluxed for four hours. The resulting solid weighed 4.2 gm. (100%) and melted at 192-193°C. A mixed melting point with an authentic sample of N, N-diphenylurea (EK 4979) was not depressed.

N, N-Diphenylmethylisourea (alkoxide catalysis)

To a solution of 0.28 gm. (0.005 mole) of potassium hydroxide in 20 ml. of methanol was added 0.97 gm. (0.005 mole) of diphenylcyanamide. The resulting solution was refluxed for two hours. After cooling, the solution was diluted with 100 ml. of water. An oil separated and crystallized after vigorous stirring. The product was found to be extremely soluble in all the common organic solvents as well as in 10% hydrochloric acid solution. Purification was achieved by dissolving it in excess acetone, then adding water dropwise while stirring at high speed. By this means 1.0 gm. (88%) of long fine needles was obtained melting at 77-78°C. Calc. for $C_{14}H_{14}ON_2$: C, 74.30; H, 6.24; $-OCH_3$, 13.71; N, 12.40%. Found: C, 74.55; H, 6.20; $-OCH_3$, 13.97; N, 12.57%.

The same product was obtained (82%) when 0.23 gm. (0.01 gm-atom) of sodium metal was used in place of potassium hydroxide.

N, N-Diphenylmethylisourea (proton catalysis)

A solution of 1.94 gm. (0.01 mole) of diphenylcyanamide in 60 ml. of methanol was saturated with dry hydrogen chloride at 6°C. After standing 80 days at 10°C., 0.08 gm. (3.5%) of N, N-diphenylmethylisourea was isolated from the solution.

N, N-Diphenylethylisourea (alkoxide catalysis)

To a solution of 2.8 gm. (0.05 mole) of potassium hydroxide in 50 ml. of absolute ethanol was added 9.7 gm. (0.05 mole) of diphenylcyanamide and the resulting solution refluxed for two hours. The reaction mixture was cooled, diluted with 150 ml. of water, and extracted with four 25 ml. portions of ether. The ether extract was treated with an excess of concentrated hydrochloric acid and the resulting precipitate taken up in 75 ml. of water. This aqueous solution was made alkaline with 20% potassium hydroxide solution. The resulting precipitate was purified by crystallization from acetone–water as described above, yielding 9.0 gm. (75%) of white crystals melting at 55-56°C. Calc. for $C_{15}H_{16}ON_2$: C, 74.96; H, 6.72; $-OC_2H_5$, 18.76%. Found: C, 74.92; H, 6.70; $-OC_2H_5$, 18.69%.

N, N-Di-p-chlorophenylurea

A suspension of 1.05 gm. (0.004 mole) of dichlorodiphenylcyanamide in 15 ml. of 6 *N* aqueous sodium hydroxide solution was refluxed for 17 hr. Bumping became so severe that the refluxing had to be stopped. The suspension was left standing at room temperature for an additional 10 days. A white crystalline solid, which melted at 220-222°C., weight 1.1 gm. (98%), was filtered off. Crystallization from ethanol and acetone–water did not raise the melting point. Calc. for $C_{13}H_{10}ON_2Cl_2$: C, 55.52; H, 3.58; N, 9.97%. Found: C, 57.20; H, 3.42; N, 10.28%.

p, p'-Dichlorodiphenylamine

A solution of 1.32 gm. (0.005 mole) of *p, p'*-dichlorodiphenylcyanamide in 50 ml. of 20% methanolic potassium hydroxide was refluxed for 24 hr. After cooling and diluting with 100 ml. of water a white precipitate was obtained which was crystallized from petroleum ether (65-110°C.). In this manner 1.03 gm. (87%) of purified material was obtained, melting at 77-78°C. A mixed melting point with an authentic sample of *p, p'*-dichlorodiphenylamine was not depressed.

N, N-Di-p-chlorophenylmethylisourea

A solution of 2.1 gm. (0.008 mole) of *p, p'*-dichlorodiphenylcyanamide in 15 ml. of methanol containing 0.56 gm. (0.01 mole) of potassium hydroxide was refluxed for two hours. The solution was cooled and diluted with 50 ml. of water. The crystalline precipitate was filtered off and crystallized four times from ethanol, yielding 1.7 gm. (72%) of purified product melting at 109-110°C. Calc. for $C_{14}H_{12}ON_2Cl_2$; C, 56.95; H, 4.08; N, 9.50; Cl, 24.04; $-OCH_3$, 10.52%. Found: C, 57.05; H, 4.28; N, 9.41; Cl, 24.14; $-OCH_3$, 10.49%.

Pyrolysis of N, N-Diphenylmethylisourea

After heating 0.9 gm. (0.004 mole) of N, N-diphenylmethylisourea in a pyrex test tube at 140-160°C. for four hours, the reaction mass was treated with 5 ml. of ether. Immediate crystallization resulted. The crystals weighed 0.1 gm. (12%) and melted at 192-193°C. after crystallization from methanol. A mixed melting point with N, N-diphenylurea showed no depression.

N, N-Diphenyl-N'-methylurea

This compound was prepared for comparison with N, N-diphenylmethylisourea in the early stages of the investigation.

A solution of 0.46 gm. (0.002 mole) of diphenylcarbamyl chloride (E.K. 801) in 25 ml. anhydrous ether was treated with excess methylamine which was distilled from a 33% aqueous solution. After one hour the reaction was stopped, the solid material filtered off, and the ether evaporated to give 0.6 gm. of crude product, which melted at 167-168°C. The crude material was purified by crystallization from methanol–water and ethanol–water giving 0.35 gm. (78%) of product melting at 173°C. Calc. for $C_{14}H_{14}ON_2$: C, 74.30; H, 6.24; N, 12.40%. Found: C, 74.40; H, 6.20; N, 12.61%.

Acknowledgments

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References

1. BASTERFIELD, S. and POWELL, E. C. Can. J. Research, 1: 261. 1929.
2. VON BRAUN, J. Ber. 33: 1450. 1900.
3. VON BRAUN, J. Ber. 33: 2724. 1900.
4. VON BRAUN, J. Ber. 33: 2725. 1900.
5. VON BRAUN, J. and SCHWARZ, R. Ber. 36: 3662. 1903.
6. BROWNING, H. C., FRASER, F. C., SHAPIRO, S. K., GLICKMAN, I., and DUBRÔLE, M. Can. J. Research, D, 26: 282. 1948.
7. CHAPMAN, A. W. J. Chem. Soc. 2461. 1930.
8. CLAUS, A. and SHAARE, H. Ber. 15: 1286. 1882.
9. HOFMANN, A. W. Ber. 18: 3219. 1885.
10. KURZER, F. J. Chem. Soc. 3035. 1949.
11. KURZER, F. J. Chem. Soc. 3032. 1949.
12. MARSHALL, E. K. and ACREE, S. F. Am. Chem. J. 49: 127. 1913.
13. MCKEE, R. H. Am. Chem. J. 26: 209. 1901.
14. MCKEE, R. H. Am. Chem. J. 36: 209. 1906.
15. MCKEE, R. H. Am. Chem. J. 42: 1. 1909.
16. MICHLER, W. Ber. 9: 397. 1876.
17. NEF, J. V. Ann. 287: 280. 1895.
18. PINNER, A. and KLEIN, FR. Ber. 10: 1889. 1877.
19. STIEGLITZ, J. and MCKEE, R. H. Ber. 32: 1494. 1899.
20. STIEGLITZ, J. and MCKEE, R. H. Ber. 33: 1517. 1900.
21. STIEGLITZ, J. Am. Chem. J. 39: 29. 1908.
22. STIEGLITZ, J. Am. Chem. J. 39: 166. 1908.
23. STIEGLITZ, J. J. Am. Chem. Soc. 32: 221. 1910.
24. STIEGLITZ, J. J. Am. Chem. Soc. 34: 1687. 1912.
25. STIEGLITZ, J. J. Am. Chem. Soc. 35: 1774. 1913.
26. WERNER, E. A. Chem. News, 65: 249. 1892.

THE ADDITION COMPOUNDS OF SILICON TETRACHLORIDE WITH ALIPHATIC AMINES[1]

By W. R. Trost

Abstract

Some diaminate and tetraminate addition compounds of silicon tetrachloride with butylamine, diethylamine, piperidine, triethylamine, and tributylamine have been prepared and analyzed. The tetraminates begin to decompose at temperatures above and below 200° C. The thermal stabilities and the products of the decompositions of these compounds indicate that coordinate bonds, and hydrogen bonds when possible, are involved in the formation of the tetraminates of silicon tetrachloride.

Addition compounds between silicon tetrachloride and aromatic amines have been reported (2) in which the silicon tetrachloride molecule adds one, two, or four amine groups to form stable substances. Coordinate bonds were considered to be principally responsible for the stability of these addition compounds. From this point of view, it was necessary to assume the presence of orbitals in silicon tetrachloride that could accept electron-pairs. These orbitals, to be associated with the silicon tetrachloride molecule as a whole, were assumed to be derived from $4s$, $3d$, $4p$ atomic orbitals. Accordingly the amines would be expected to add as an envelope around the silicon tetrachloride molecule. Aliphatic amines also form addition compounds with silicon tetrachloride. These should be the stabler substances, as the aliphatic amines are much stronger bases than the aromatic amines and would therefore be supposed to be able to form stronger coordinate bonds.

Experimental

The tetraminates of silicon tetrachloride with butylamine, piperidine, diethylamine, triethylamine, and tributylamine were prepared by adding 10 ml. of 1.74 M silicon tetrachloride in ether solution to 100 ml. of the distilled, chilled, pure amine. The mixture was diluted with ether and the solid filtered and washed with ether until free of unbound amine, when it was dried *in vacuo*. The yields were quantitative.

The dry powders were analyzed for amine, chlorine, and silicon. The analytical results are reported in Table I. The amines were determined by back titrating the acid fixed condensate from the boiling alkaline solution of the solid. The chlorine was separately measured in neutral solution with standard silver nitrate, after the volatile amine had been removed by distillation from basic solution, and the silica had been precipitated by acid. Silicon was calculated from silica, recovered as the ash in the ignition of the tetraminate.

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Contribution from the Department of Chemistry, Dalhousie University, Halifax, Canada, with financial assistance from the National Research Council of Canada.

TABLE I

RESULTS OF THE ANALYSES OF THE ADDITION COMPOUNDS

Compound	Experimental			Calculated		
	% Si	% Cl	% Amine	% Si	% Cl	% Amine
$SiCl_4(Et_3N)_4$	4.75	23.6	69.6	4.88	24.7	70.5
$SiCl_4(Bu_3N)_4$	3.16		81.0	3.09		81.3
$SiCl_4(Bu_3N)_2$	5.15		68.1	5.19		68.5
$SiCl_4(Et_2NH)_4$	6.07	30.3	63.2	6.08	30.7	63.3
$SiCl_4((CH_2)_5NH)_4$	5.50	27.2	65.5	5.51	27.8	66.6
$SiCl_4(BuNH_2)_4$	6.01	31.0	63.8	6.08	30.7	63.3

Tributylamine is not volatile. Therefore, in analyses for this amine, as it was possible, the method applied before to the aromatic amines was used (2).

The aliphatic tetraminates, particularly those derived from tertiary amines, are relatively inert to water. They are not hygroscopic and must be refluxed in such solvents as boiling dilute alkali to obtain complete decompositions. They are insoluble in common organic liquids, including amines.

Thermal Stability

The aliphatic tetraminates decomposed without melting when kept at high temperatures. The recognizable products of the early stages of the decomposition were volatile colorless liquids and white crystalline solids, at room temperature. The crystals were tentatively identified as the corresponding amine hydrochloride by their melting points, and because of positive qualitative tests for ionically bound chlorine. The volatile liquids neutralized acids and had the proper amine odors and were therefore taken to be the corresponding amines. The lowest temperatures at which the decompositions of these substances were observed to begin are compared with the basic dissociation constants (1) of the relevant amines in Table II.

TABLE II

THE THERMAL STABILITY OF THE ADDITION COMPOUNDS COMPARED WITH THE BASIC STRENGTH OF THE CORRESPONDING AMINES

Compound	Decomposition temperature, °C.	Amine	K_B (1)
$SiCl_4(Et_3N)_4$	210	Triethylamine	6.4×10^{-4}
$SiCl_4(Bu_3N)_4$	185	Tributylamine	8.5×10^{-5}
$SiCl_4(Me_2Aniline)_4$ (2)	100	Dimethylaniline	1×10^{-9}
$SiCl_4((CH_2)_5NH)_4$	220	Piperidine	1.6×10^{-3}
$SiCl_4(Et_2NH)_4$	215	Diethylamine	1.26×10^{-3}
$SiCl_4(BuNH_2)_4$	215	Butylamine	4.09×10^{-4}
$SiCl_4(Aniline)_4$ (2)	185	Aniline	4.6×10^{-10}

1. $SiCl_4(Et_3N)_4$

The tertiary amine tetraminates were refluxed in excess amine and inert solvent at 80° C., then washed, and dried *in vacuo* again, prior to their thermal decompositions.

The triethylamine compound began to decompose at about 210° C. The solid remained dry, but soon started to char. Triethylamine was the principal product at first, but white crystals also began to collect, condensing from the gas phase away from the reaction zone. On further heating at higher temperatures the amine disappeared, the charring solid turned black, the white crystals continued to gather, and other volatile products appeared. The black ash, burnt in air, transformed to silica.

2. $SiCl_4(Bu_3N)_4$

The tributylamine tetraminate is a pale yellow powder that began to decompose at about 185°, becoming brown and sticky with liquid amine that was only partly volatile. The first stages of the decomposition were followed at higher temperatures by charring in the dry solid and the formation of amine chlorides.

3. $SiCl_4(Et_2NH)_4$

The white powder started to decompose without charring at about 215°, releasing some amine, while amine hydrochloride also formed as white crystals that had condensed from the vapors. Charring in the solid began only at higher temperatures.

4. $SiCl_4((CH_2)_5NH)_4$

Decomposition, without charring of the white powder, began at about 220°. The principal volatile product was the amine hydrochloride, although some free amine might have been present. When charring began at higher temperatures, the solid had already lost most of its chlorine.

5. $SiCl_4(BuNH_2)_4$

Decomposition of the primary amine addition compound began at about 215° with the appearance of a colorless liquid that clotted the white powder and froze to form a white solid that corresponded to butylammonium chloride. No free amine was observed.

Aliphatic Diaminates

What were presumed to be the diaminates of the five aliphatic amines were observed, but only the tributylamine diaminate was analyzed (Table I). It is a brown solid that reacts quickly with moist air, water, or excess amine. It was recovered from silicon tetrachloride mixed with tributylamine in the ratio of 15/1. The other addition compounds, assumed to be diaminates, were all yellow in color, and generally reactive.

Discussion

The decomposition of the tetraminates of the tertiary amines apparently begins with the liberation of the amine from the addition compound. The temperature at which decomposition begins is lowest for the weakest base, dimethylaniline (100° C.), and highest for the strongest base, triethylaniline (210°). These comparisons are shown in Table II. The strength of coordinate bonds would also be expected to increase with the basicity of the donor group. Therefore the relative stabilities of the tertiary amine addition compounds will be associated with the strengths of the coordinate bonds between the amine and the silicon tetrachloride.

The compounds formed from primary amines decompose initially without charring, with the formation of the appropriate amine hydrochloride. The temperature required for decomposition is higher for the stronger base, butylamine (215° C.) than for the weaker base, aniline (185° C.). But the first steps in the decomposition must now include the elimination of hydrogen chloride from the addition compound. If this precedes, or is simultaneous with, the dissociation of free amine from the addition compound, rather good mechanistic evidence for the presence of hydrogen bonds in the addition compounds would be provided. No free amine was detected in the decomposition of the primary amine compounds. Again, the aniline tetraminate is thermally much stabler (185° C.) than the tetraminate of the stronger base, dimethylaniline (100° C.). It therefore appears plausible, and perhaps necessary, to assume that, in addition to coordinate bonds, these addition compounds are stabilized by hydrogen bonds to the chlorine atoms in silicon tetrachloride, when the amine nitrogen is hydrogenated.

As the thermal decompositions of the tertiary amine compounds and of the primary amine compounds are different chemical reactions, the decomposition temperatures of these compounds do not properly compare their relative stabilities, for a limit to the thermal stability of the compounds is the temperature at which the disruptive chemical reaction, the formation of amine hydrochlorides, occurs, and not simply the dissociation of the addition compounds. It might be expected that heats of formation would better show the stability of the primary amine compounds, relative to the tertiary amine compounds, than do the decomposition temperatures.

References

1. HALL, N. F. and SPRINKLE, M. R. J. Am. Chem. Soc. 54: 3469. 1932.
2. TROST, W. R. Can. J. Chem. 52: 877. 1951.

THE MERCAPTOLYSIS OF GLUCOSE AND GALACTOSE PENTAACETATES[1]

By R. U. Lemieux

Abstract

The mercaptolysis of the α-and β-pentaacetyl derivatives of glucopyranose and galactopyranose at 0°C. with zinc chloride catalyst was studied. The β-pentaacetates were rapidly transformed to the corresponding ethyl tetraacetyl-1-thio-β-D-glycosides in excellent yields; however, the α-anomers were highly resistant to the mercaptolytic conditions. The results are interpreted on the basis of a participation by the C_2-acetoxy group in the replacements at the lactol carbon atom of the β-pentaacetates with the intermediate formation of a resonance stabilized cyclic carbonium ion.

Brigl and Schinle (8) first studied the interaction of acylated cyclic sugars with ethyl mercaptan in the presence of an acid catalyst. Hydrochloric acid was used as catalyst and the too rapid deacetylation of the pentaacetyl glucoses led to the use of the pentabenzoyl derivatives. Under the conditions used, the pentabenzoyl glucopyranoses remained unchanged. In 1934, Wolfrom and Thompson (36) modified the procedure by substituting anhydrous zinc chloride as catalyst and by operating in the presence of either anhydrous sodium sulphate or Drierite (anhydrous calcium sulphate) as drying agents. In this manner, the acid hydrolysis of the acetyl derivatives was minimized. They showed that the treatment of the fully acetylated cyclic form of a ketose with ethyl mercaptan and zinc chloride led to the replacement of the acetoxy group at the lactol carbon atom by the ethylthio group. Furthermore, they showed that the acetyl derivatives of *aldehydo-* and *keto*-sugars yielded the diethyl thioacetals without loss of acetoxy group. These conditions for mercaptolysis therefore comprised a valuable method for the differentiation of cyclic and open-chain acetyl derivatives of ketoses (36). However, statements (27, p. 154) to the contrary not withstanding, there appears to be no report of applications of this mercaptolysis procedure to fully acetylated cyclic aldoses.

This communication deals with the results obtained when the α- and β-pentaacetyl derivatives of D-glucopyranose and D-galactopyranose were interacted with ethyl mercaptan and zinc chloride. Ethyl tetraacetyl-1-thio-β-D-glucopyranoside was obtained in 71.2% yield by the mercaptolysis of pentaacetyl-β-D-glucopyranose. Mercaptolysis of pentaacetyl-β-D-galactopyranose yielded ethyl tetraacetyl-1-thio-β-D-galactopyranoside in 75.6% yield. However,

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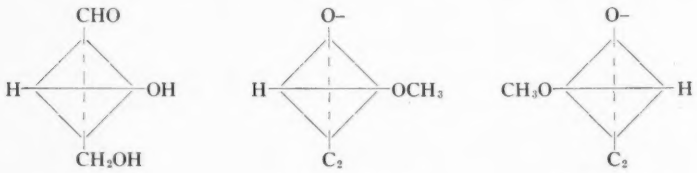
the mercaptolysis of the α-pentaacetyl compounds was not a practical means for the preparation of 1-thioglycosides. Schneider, Sepp, and Stiehler (32) prepared ethyl tetraacetyl-1-thio-β-D-glucopyranoside in 19% yield through the interaction of acetobromoglucose with potassium mercaptide. Ethyl 1-thio-α-D-glucopyranoside was obtained in 20% yield by treating glucose diethyl mercaptal with 22% hydrochloric acid (6, 26). Fried and Walz (9) have prepared the anomeric ethyl tetraacetyl-1-thio-D-manno- and galactopyranosides in low yield by the prolonged treatment of the sugar with ethyl mercaptan and concentrated hydrochloric acid followed by acetylation.

The melting point, 82.5-83°C. and $[\alpha]_D -28°$ found for ethyl tetraacetyl-1-thio-β-D-glucopyranoside were higher than the values, m.p. 78-79°C. (32), $[\alpha]_D -25.6°$ (25), previously reported. However, Schneider, Sepp, and Stiehler (32) have reported that some of their preparations melted at 82°C. The identity of the compound was established by deacetylation to the thioglucoside and reductive desulphurization to tetraacetyl-1,5-anhydro-D-sorbitol. The physical constants found for the ethyl tetraacetyl-1-thio-β-D-galactopyranoside and its deacetylated derivative were in agreement with those recently reported by Fried and Walz (9).

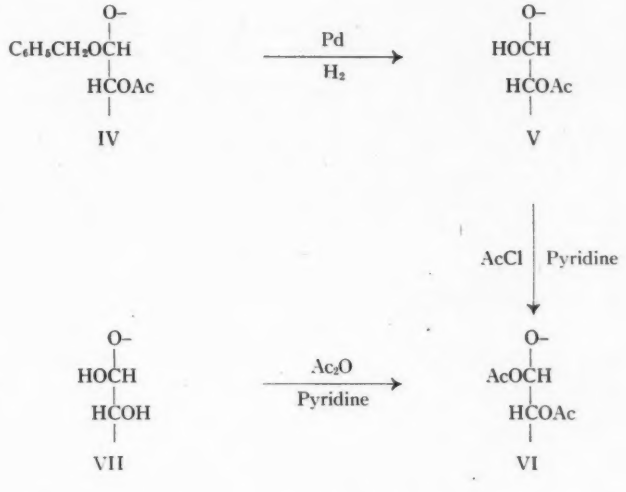
Hudson's Rules of Isorotation (15) state that for an α, β-pair of anomers the more dextrorotatory member is designated either α-D or β-L and the more levorotatory member is termed either β-D or α-L. As pointed out by Hudson (16, p. 17) these rules are no longer of a wholly empirical nature and at least for the methyl glycosides of many sugars (20) the rules have consistently allocated, to each of the α-and β-series, compounds of like configuration for the lactol carbon atoms. Ballou, Roseman, and Link (1) recently have reviewed the basis for the allocation of anomeric configurations. However, these authors overlooked the important and decisive argument for the configuration of methyl β-D-glucopyranoside advanced by Hudson (16, p. 17) based on the methanolysis of D-glucosan $<1, 5>$ α $<1, 2>$ (13). Thus, for the many methyl glycosides studied (20), it seems clear that the configurations of the lactol carbon atoms relative to the configuration of D-glyceraldehyde (I) are as shown



I α – D or β – L II β – D or α – L III

in II and III. These configurations are homologous to the accepted configurations of the α- and β-glucopyranoses (4). Ballou, Roseman, and Link (1) have

converted some acetylated benzyl glycosides to the corresponding 1-hydroxy polyacetyl aldoses by catalytic hydrogenolysis and have thus related the anomeric configurations of these compounds. Thus, benzyl tetraacetyl-β-D-glucopyranoside (IV) was converted to 2,3,4,6-tetraacetyl-β-D-glucopyranose (V). We have acetylated the latter compound (12) with acetyl chloride in benzene-pyridine to pentaacetyl-β-D-glucopyranose (VI) in 70% yield. Since this



acetylation would be expected not to affect the configuration of the lactol carbon atoms, the relative configurations of V and VI would be as shown. It has long been observed (2) that the low temperature acetylation of a free sugar by acetic anhydride in pyridine solution yields a polyacetyl derivative of the same anomeric configuration. Thus, the acetylation of β-D-glucopyranose (VII) under these conditions yields the β-D-pentaacetyl derivative (17). Considering the fact that these chemical transformations infer the same relative configurations for the substances IV to VII and that these conclusions are in accord with the predictions based on Hudson's Rules of Isorotation, there can be little doubt that the structure of pentaacetyl-β-D-glucopyranose (VI) is as shown. Furthermore, considering the success of the Rules of Isorotation in the correlation of the anomeric configurations of several methyl glycosides (20), there can be little doubt about the configurations of any pair of truly anomeric aldoses or their simple derivatives as allocated on the basis of these rules.

The treatment of pentaacetyl-β-D-glucopyranose with 10% zinc chloride in dry ethyl mercaptan at 0°C. for 12 hr. gave ethyl tetraacetyl-1-thio-β-D-glucopyranoside in a minimum yield of 71.2%. The residual sirups could not be

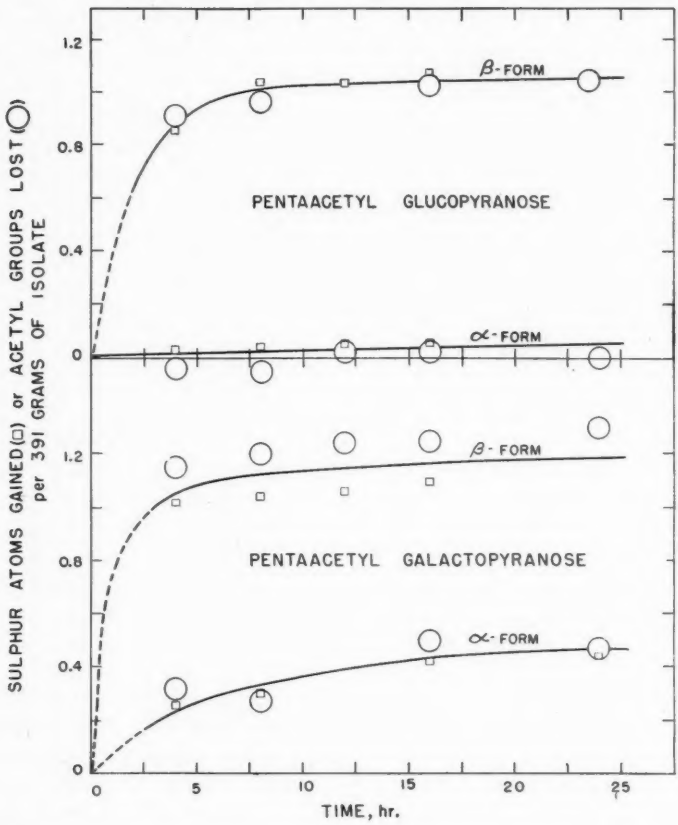
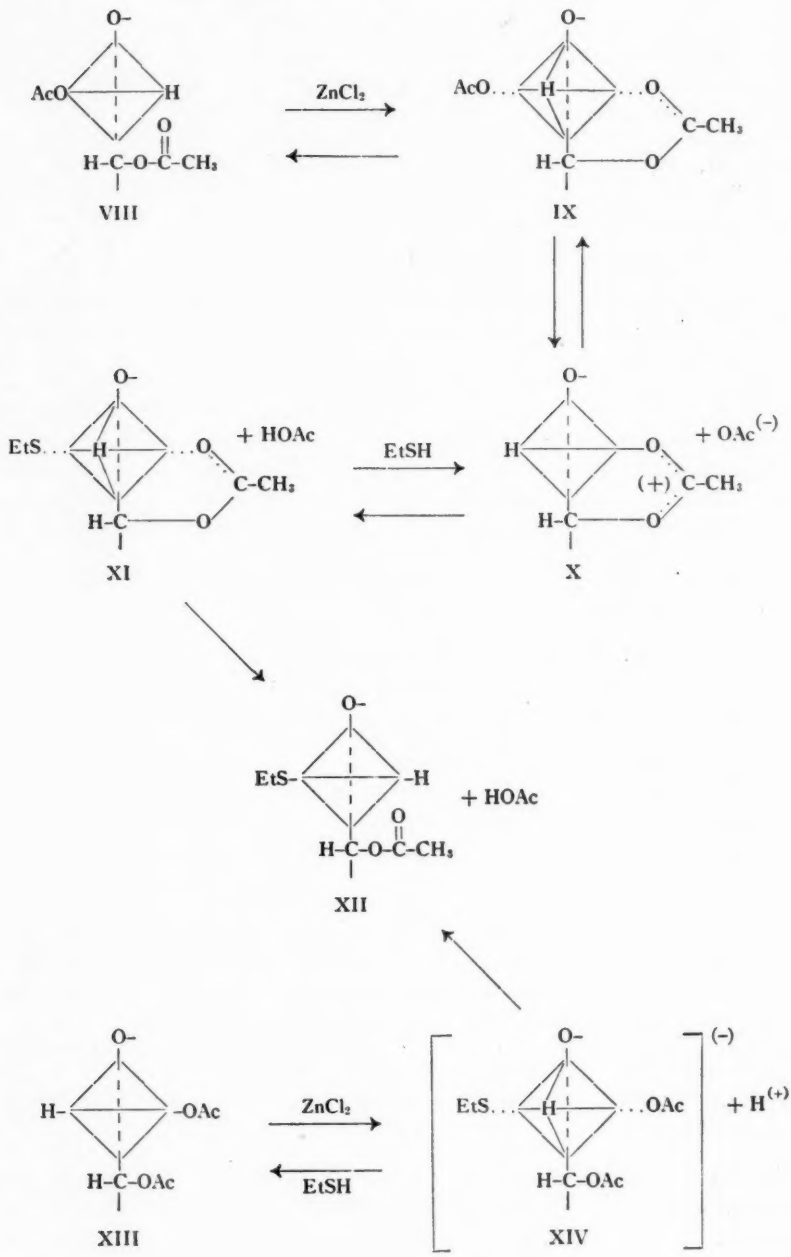


FIG. 1. *The rates of mercaptolysis of the anomeric pentaacetates of* D-*glucose and* D-*galactose in zinc chloride–ethyl mercaptan solution at 0°C.*

made to yield starting material. A similar treatment of pentaacetyl-α-D-glucopyranose brought about little reaction and the starting material was recovered nearly quantitatively. However, by operating at room temperature and extending the reaction time to 42 hr., the α-anomer was transformed to an isolable amount of ethyl tetraacetyl-1-thio-β-D-glucopyranoside but a large quantity of the starting material remained unaffected. In order to more definitely establish this marked difference in reactivity between the two anomers toward the mercaptolytic conditions, the rates of mercaptolysis were determined at 0°C. At various time intervals, the glucose derivatives in an aliquot of the reaction mixture were isolated and the product was analyzed for acetyl group and sulphur content. Although it was clear that the replacement of the C1-acetoxy group by the ethylthio group was accompanied by other reactions, the curves in Fig. 1 were obtained by plotting against time both the number of acetyl groups lost and the number of sulphur atoms gained calculated on the assumption that both the starting material and the reaction products possessed a molecular weight of 391. The figure 391 is purely arbitrary and was chosen because of its close agreement with the molecular weights of both the starting materials, 390, and ethyl tetraacetyl-1-thio-β-D-aldohexopyranoside, 392. Thus, Fig. 1 shows clearly the great difference in reactivity between the anomeric glucose pentaacetates under the reaction conditions used and it is clear that the main reaction involved the substitution of acetoxy groups by ethylthio groups.

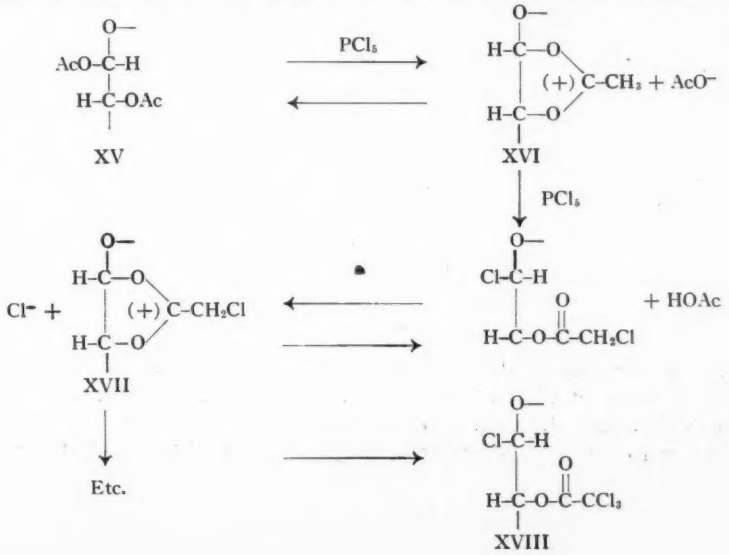
Therefore, the mercaptolytic replacement of the C1-acetoxy group of pentaacetyl-β-D-glucopyranose, which contains the C1- and C2-acetoxy groups in *trans* relationship, proceeded with retention of configuration while the C1-acetoxy bond of pentaacetyl-α-D-glucopyranose, which contains the C1- and C2-acetoxy groups in *cis* relationship, underwent mercaptolysis with inversion of the configuration of the lactol carbon. The stereochemical routes of the reactions are therefore reminiscent of the findings by Winstein and Buckles (35) on the role of neighboring groups in replacement reactions. Furthermore, these authors pointed out that the participation of a neighboring group greatly facilitates a replacement reaction. Thus, they showed that under conditions more drastic than those needed to bring about reaction of the *trans*-1-acetoxy-2-chlorocyclohexane with silver acetate, the *cis*-isomer remained completely unchanged. The results led the authors to state, "The *trans*-acetoxy group seems to be able to supply some driving force for the reaction and the *cis*-acetoxy group is essentially unable to do this." Similar effects have been noted for the replacement of mesyloxy (23) and tosyloxy (3) groups situated in ring structures and the activating effect of a participating neighboring group appears widespread.

It was thus apparent that the mechanisms of the main reactions for the mercaptolyses of the anomeric glucose pentaacetates could be formulated as follows.

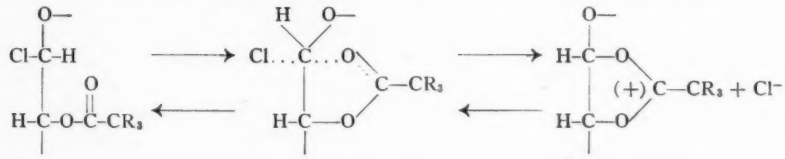

AcO
H
O
H–C–O–C–CH₃
ZnCl₂
AcO
H
O
C–CH₃
H–C
O
VIII
IX
EtS
H
O
C–CH₃
H–C
O
+ HOAc
EtSH
H
O
(+) C–CH₃
H–C
O
+ OAc(–)
XI
X
EtS–
–H
H–C–O–C–CH₃
+ HOAc
XII
H–
–OAc
H–C–OAc
ZnCl₂
EtSH
EtS
H
OAc
H–C–OAc
(–)
+ H(+)
XIII
XIV

For the pentaacetyl-β-D-glucopyranose (VIII), the acid catalyzed removal of the C1-acetoxy group was likely facilitated (11, p. 131; 34, p. 216) by the backside approach of the nucleophilic C2-acetoxy group with the transition state (IX) decomposing to the resonance stabilized α-D-carbonium ion (X). The neutralization of this ion, under the prevailing acidic conditions, would proceed through the transition state (XI) with inversion of the lactol carbon atom to yield the ethyl tetraacetyl-1-thio-β-D-glucopyranoside (XII) a substance which is known (32) to be highly stable under acidic conditions. The acid catalyzed removal of the C1-acetoxy group of the pentaacetyl-α-D-glucopyranose (XIII) was probably facilitated through a backside approach by a nucleophilic group provided by the solvent, ethyl mercaptan, to yield the transition state (XIV) which decomposed to the ethyl tetraacetyl-1-thio-β-D-glucopyranoside (XII).

Isbell (19) has noted that reactions of acetohalogeno-sugars tend to yield derivatives with the C1 and C2 atoms in *trans*-relationship. The possible formation of cyclic carbonium ions of the type of structure shown for X was definitely established by Lucas, Mitchell, and Garner (22). Hurd and Holysz (18) have established the formation of 1, 2-ketal derivatives of 3, 4, 6-triacetyl-α-D-glucopyranose on the interaction of tetraacetyl-α-D-glucopyranosyl bromide with dialkylcadmium. Thus, it is sterically possible for the carbonyl oxygen of an acetoxy group situated at the C2-position of glucopyranose to interact with the lactol carbon atom. The preparation by Brigl (5) of 2-trichloroacetyl-3,4,6-triacetyl-β-D-glucopyranosyl chloride (XVIII) by the interaction of pentaacetyl-β-D-glucopyranose (XV) with phosphorus pentachloride appears best rationalized, on the basis of modern chemical theory, by the assumption that the preferential chlorination of the C2-acetoxy group is related to an activation of the C-H bonds by the formation under the reaction conditions of the cyclic carbonium ion intermediates such as XVI and XVII. Thus, the reaction is probably quite accurately formulated as follows.

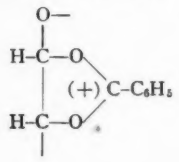


Schlubach, Stadler, and Wolf (31) have shown that tetraacetyl-β-D-glucopyranosyl chloride (30), although quite stable in benzene, ether, or carbon tetrachloride solution, rapidly mutarotated in acetonitrile or chloroform solution and the transformation was nearly complete after 96 hr. On the other hand, Hickinbottom (14) has shown that trichloroacetyl-3,4,6-triacetyl-β-D-glucopyranosyl chloride does not exhibit any appreciable anomerization in benzene or acetonitrile after 500 hr. and only a slight increase in rotation occurs in chloroform after 540 hr. This stabilizing effect may be related to the weaker nucleophilic properties of the carbonyl oxygen of the trichloroacetyl group as compared to the carbonyl oxygen of the acetyl group. That is, it is reasonable to expect that the establishment of the transition state (XIX) and the carbonium ion (XX) would be more difficult with R = Cl than R = H.



XIX XX

Thus, it is apparent that the postulation of the cyclic carbonium ion (X) as an intermediate in many replacement reactions at the lactol carbon atom of acylated glucose derivatives is not pure speculation since the idea is strongly supported by highly plausible interpretations of a variety of established experimental facts. It is noteworthy that Ballou, Roseman, and Link (1) have recently reported that, on hydrogenolysis, the acetylated benzyl glucoside, xyloside, and arabinoside in which the aglycon and C2-acetoxy groups are *trans* cleave much more easily than those with a *cis* relationship. Ness, Fletcher, and Hudson (24) have recently shown that the β-forms of both tribenzoyl-D-ribopyranosyl chloride and bromide reacted much more rapidly in 1:9-dioxane-methanol than did the corresponding α-anomers and in each case methyl tribenzoyl-β-D-ribopyranoside was the principal product formed. It was postulated that the reactions of the β-D(*trans*)-halides involved the intermediate formation of the cyclic carbonium ion (XXI).



XXI

As was found for the pentaacetyl glucoses, under the same reaction conditions, the mercaptolysis of pentaacetyl-β-D-galactopyranose proceeded much more rapidly than did the mercaptolysis of the α-anomer (see Fig. 1). Only starting material could be isolated by crystallization of the product from the

mercaptolysis of the α-anomer for 60 hr. at 0°C. On the other hand, the same treatment of the β-pentaacetate for only 12 hr. gave ethyl tetraacetyl-1-thio-β-D-galactopyranoside in high yield. It is therefore probable that the mercaptolysis of pentaacetyl-β-D-galactopyranose involved the participation of the C2-acetoxy group with the formation of a transient cyclic carbonium ion in the manner indicated above for pentaacetyl-β-D-glucopyranose.

Deacetylation of the residual sirupy materials from the mercaptolysis at room temperature of both the α- and β-pentaacetates of glucose yielded the 2-dehydroxy-2-ethylthio-D-glucose (D-mannose?) diethyl mercaptal of Brigl, Mühlschlegel, and Schinle (7).

The mercaptolysis of other anomeric pairs of sugar acetates is being studied.

Experimental

The rotations were measured at room temperature, 23-28°C., and the melting points were corrected.

Ethyl tetraacetyl-1-thio-β-D-glucopyranoside

Dry pentaacetyl-β-D-glucopyranose, 20 gm., was dissolved at 0°C. in anhydrous ethyl mercaptan, 100 ml., which contained 10 gm. of anhydrous zinc chloride. The mixture was shaken until solution was complete and left for a total of 12 hr. in ice water. The resulting mixture was poured into a stirred, saturated aqueous solution of sodium bicarbonate, 500 ml. The precipitate was collected by filtration and it was stirred with 150 ml. of boiling chloroform. The residue was collected by filtration and the chloroform filtrate was used to extract the aqueous filtrate from the first filtration. The procedure for the extraction of the precipitate and aqueous solution was repeated twice more. The combined chloroform extracts were washed with water and dried over sodium sulphate. The solvents were removed *in vacuo* to yield a colorless sirup which soon crystallized. The crystalline mass was extracted with 10 ml. of ether followed by 30 ml. of 50% ether–Skellysolve F. The air-dried material, m.p. 78-80.5°C., weighed 10.26 gm. The ethereal filtrate was evaporated to sirup which was dissolved in 20 ml. of ether and Skellysolve F was added to turbidity. After standing in the cold for 24 hr. a second crop was obtained, 4.01 gm., which melted at 79-81°C. The total crude yield was 71.2%. The two crops were combined and recrystallized from 10 ml. of ethanol to yield 13.23 gm. of product, m.p. 80.5-82°C. After several further recrystallizations from ethanol, the substance melted at 82.5-83°C. with $[\alpha]_D - 28°$ (*c*, 0.9 in chloroform). Calc. for $C_{16}H_{24}O_9S$: S, 8.16; acetyl, 45.84%. Found: S, 8.13; acetyl, 44.1%. Schneider, Sepp, and Stiehler (32) have reported the constants, m.p. 78-79°C., $[\alpha]_D^{20} - 22.27°$ (*c*, 2.6 in acetylene tetrachloride), while Pacsu and Wilson (25) have recorded $[\alpha]_D - 25.6°$ (*c*, 1.464 in chloroform) for ethyl tetraacetyl-1-thio-β-D-glucopyranoside.

When 20 gm. of pentaacetyl-β-D-glucopyranose was mercaptolyzed under the above conditions, with the exception that 15 gm. of anhydrous calcium

sulphate, "Drierite", was added to the reaction mixture and that the solution was kept at room temperature for 17 hr., the yield of crude product was only 6.90 gm. The residual sirupy material was deacetylated in methanol solution with barium methylate as catalyst in the usual manner. Extraction of the product with ether yielded a substance which after two recrystallizations from water melted at 101-102°C. with $[\alpha]_D + 2.3°$ (*c*, 1.2 in acetone). These constants are in close agreement with those recorded for the 2-dehydroxy-2-ethylthio-D-glucose (mannose?) diethyl mercaptal of Brigl, Mühlschlegel, and Schinle (7), m.p. 102°C. (little sintering at 101°C.), $[\alpha]_D + 2.27°$ (*c*, 2.2 in acetone). Calc. for $C_{12}H_{26}O_4S_3$: S, 29.1%. Found: S, 28.6%.

Dry pentaacetyl-α-D-glucopyranose, 5 gm., was dissolved in dry ethyl mercaptan (27 ml.) which contained anhydrous zinc chloride, 4 gm., and 4 gm. of anhydrous sodium sulphate. After standing at room temperature for 42 hr., the mixture was poured with stirring into 200 ml. of saturated aqueous sodium bicarbonate solution. The resulting mixture was extracted with chloroform in the manner described above. The extract was reduced to a sirup which was treated with about 25 ml. of Skellysolve F and the solvent was removed by blowing. After this procedure was repeated three times, crystallization soon began. Ether was added to dissolve the sirupy phase and Skellysolve F was added to turbidity. The crystals which formed weighed 1.40 gm. and melted at 109-111°C. and were found to be essentially pure pentaacetyl-α-D-glucopyranose. The mother liquor was reduced to yield 3.25 gm. of a pale-yellow sirup, $[\alpha]_D + 28°$ (chloroform). This substance was dissolved in ether and Skellysolve C was added to turbidity. After seeding with ethyl tetraacetyl-1-thio-β-D- glucopyranoside, the solution was allowed to stand overnight at 4°C. Crystals, 0.73 gm., $[\alpha]_D - 0.1°$ (chloroform), were deposited which, after two recrystallizations from ethanol, yielded 275 mgm. of material, m.p. 81-82°C., $[\alpha]_D - 25.8°$ (chloroform). This melting point was not depressed on admixture with ethyl tetraacetyl-1-thio-β-D-glucopyranoside. The main mother liquors were reduced to sirup which was dissolved in ether and Skellysolve F was added to turbidity. After standing at 4°C. for some time 0.15 gm. of pentaacetyl-α-D-glucopyranose was deposited. The mother liquors were taken to a dry sirup which was deacetylated in methanol solution with barium methylate catalyst. Extraction of the product with ether yielded a crystalline material which after recrystallization from ethanol–water melted at 100-101°C. with $[\alpha]_D + 1.4°$ (*c*, 1 in acetone) and this melting point was not depressed on admixture with the above described 2-dehydroxy-2-ethylthio-D-glucose (mannose?) diethyl mercaptal. The yield of recrystallized product was 110 mgm. The sirup which remained after the above ether extraction was dissolved in water and treated with yeast for one day. After removal of the yeast cells, the solution was concentrated *in vacuo* to a dry sirup, 230 mgm., which was acetylated with acetic anhydride and sodium acetate. The product, $[\alpha]_D + 40°$ (chloroform) has not crystallized. It was therefore apparent that little, if any, ethyl tetraacetyl-1-thio-α-D-glucopyranoside, $[\alpha]_D + 194°$ (chloroform) (25), was present in the reaction product.

Deacetylation of the ethyl tetraacetyl-1-thio-β-D-glucopyranoside in methanol solution and barium methylate catalyst (9) yielded a product which after three recrystallizations from ethanol – ethyl acetate melted at 99.5-100.5°C. with $[\alpha]_D - 57°$ (*c*, 0.9 in water). Schneider, Sepp, and Stiehler (32) have reported ethyl-1-thio-β-D-glucopyranoside to melt at 99-100°C. with $[\alpha]_D^{25} - 55.14°$ (*c*, 4 in water).

1, 5-Anhydro-D-sorbitol (polygalitol)

Ethyl tetraacetyl-1-thio-β-D-glucopyranoside, 1.0 gm., was dissolved in 70% aqueous ethanol and 20 ml. of settled Raney nickel catalyst was added. After refluxing for five hours, the catalyst was removed by filtration and washed three times with hot ethanol. The combined filtrates were reduced *in vacuo* to a sirup which crystallized after 10 days. After recrystallization from ether-Skellysolve F the yield was 500 mgm. of a substance, m.p. 55-65°C., which was recrystallized from water followed by recrystallization from ether-Skellysolve C. The nicely crystalline product melted at 65-66°C. with $[\alpha]_D + 40°$ (*c*, 0.6 in chloroform). Calc. for $C_{14}H_{20}O_9$: acetyl, 51.81%. Found: acetyl, 51.5%. Richtmyer and Hudson (29) have definitely established the dimorphous character of tetraacetyl polygalitol, $[\alpha]_D^{20} + 38.9°$ (*c*, 2 in chloroform), with melting points at 65-67°C. and 73-74°C.

A solution of tetraacetyl polygalitol, 150 mgm., in 15 ml. of dry methanol was saturated with ammonia. After four hours at room temperature, the solution was reduced to a sirup which soon crystallized. Recrystallization from dry methanol gave 60 mgm., 81% yield, of a substance, m.p. 141-141.5°C., $[\alpha]_D + 43°$ (*c*, 0.5 in water). Polygalitol is reported (28) to melt at 141-142°C. with $[\alpha]_D + 42.4°$ (*c*, 2 in water).

Ethyl Tetraacetyl-1-thio-β-D-galactopyranoside

Dry pentaacetyl-β-D-galactopyranose, 5 gm., was dissolved at 0°C. in 25 ml. of dry ethyl mercaptan which contained 2.5 gm. of anhydrous zinc chloride and the reaction mixture was kept in ice water for 12 hr. The reaction product was isolated as described above and the sirupy material soon crystallized. The material was dissolved in 15 ml. of ether and 5 ml. of Skellysolve F and the solution was cooled and seeded. After crystallization appeared complete, 10 ml. of Skellysolve F was added and the mixture was kept at 4°C. for two hours. Filtration gave 3.78 gm. of material, m.p. 71.5-73°C., 76.5% yield. After two recrystallizations from ether-Skellysolve F, the material melted at 74.5-75°C. with $[\alpha]_D - 8.5°$ (*c*, 2.3 in chloroform). Calc. for $C_{16}H_{24}O_9S$: acetyl, 43.64%. Found: acetyl, 43.8%. Fried and Walz (9) have reported the constants, m.p. 74-75°C., $[\alpha]_D^{26} - 8.0°$ (*c*, 2.1 in chloroform) for ethyl tetraacetyl-1-thio-β-D-galactopyranoside.

The substance, 500 mgm., was deacetylated in methanol solution with barium methylate catalyst to yield a product which was recrystallized from ethanol – ethyl acetate. The yield was 200 mgm., m.p. 122-122.5°C., $[\alpha]_D -$

23.5° (*c*, 1.1 in water). Fried and Walz reported (9) the constants, m.p. 121.5-122.5°C., $[\alpha]_D^{26} - 23.5°$ (*c*, 0.89 in water), for ethyl 1-thio-β-D-galactopyranoside.

Dry pentaacetyl-α-D-galactopyranose, 5 gm., was subjected to mercaptolysis under the conditions described for the β-anomer except that the reaction time was extended to 60 hr. The sirupy reaction product was isolated in the usual way and was freed of chloroform by the addition of Skellysolve C followed by removal of the solvents *in vacuo*. This procedure was repeated several times. The resulting sirup was dissolved in ether and Skellysolve C was added to turbidity. After cooling, the solution was seeded with pentaacetyl-α-D-galactopyranose and left at 4°C. for 14 days. The crystals, 0.73 gm., which deposited were washed with a little ether and recrystallized from alcohol. The melting point was 91-93.5°C. with $[\alpha]_D + 98°$ (*c*, 1.8 in chloroform) and this melting point was not depressed on admixture with the starting material, m.p. 94-95°C., $[\alpha]_D + 105°$ (*c*, 1 in chloroform). The residual sirup, $[\alpha]_D + 22°$ (*c*, 3.6 in chloroform), could not be made to crystallize.

Rates of Mercaptolysis

Ethyl mercaptan, 90 ml., dried over anhydrous sodium sulphate, was added to 10.1 gm. of anhydrous zinc chloride at 0°C. and the mixture was shaken until solution was complete. At zero time, 15 ml. aliquots of the solution were added at 0°C. to 3.0 gm. samples of the sugar acetates. The glass-stoppered flasks were shaken until solution appeared complete. The contents of each flask were then distributed by means of a syringe as 2 ml. aliquots into dry test tubes. At the various time intervals, the content of a test tube was poured into 25 ml. of saturated sodium bicarbonate solution. After stirring until reaction appeared complete, the white precipitate was collected by filtration and stirred with 15 ml. of boiling chloroform. The mixture was filtered and the filtrate was collected with the aqueous filtrate of the first filtration. After shaking, the chloroform layer was washed twice with water. This extraction procedure was repeated two more times using the same equipment and wash waters. The chloroform extracts were combined, dried by filtration through dry filter paper, and reduced *in vacuo* to a sirup. The sirup was first dried at 65°C. under partial vacuum in a stream of air, then at 10^{-2} mm. for two days at room temperature. The acetyl group contents were determined at 0°C. by the procedure developed by Kunz (21) but on a semimicro scale. The sulphur estimations were by the volumetric dry combustion technique (33).

Acetylation of 2, 3, 4, 6-Tetraacetyl-β-D-glucopyranose

Acetyl chloride, 0.28 gm., was dissolved in 20 ml. of dry benzene and 5 ml. of dry pyridine was added at 0°C. 2,3,4,6-Tetraacetyl-β-D-glucopyranose (10), 1 gm., was added and the mixture was shaken for 15 min. at 0°C., and for 1.5 hr. at room temperature. Ice water, 100 ml., was added and, after vigorous shaking, the benzene layer was separated and washed with water. The dried benzene solution was reduced in a stream of air to a crystalline material which

was recrystallized from ethanol. The yield was 0.75 gm., 70%, m.p. 131-132°C. The melting point was not depressed on admixture with penta-acetyl-β-D-glucopyranose, m.p. 131-132°C.

Acknowledgments

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References

1. BALLOU, C. E., ROSEMAN, S., and LINK, K. P. J. Am. Chem. Soc. 73: 1140. 1951.
2. BEHREND, R. and ROTH, P. Ann. 331: 359. 1904.
3. BELL, D. J. and WILLIAMSON, S. J. Chem. Soc. 1196. 1938.
4. BOESEKEN, J. and COUVERT, H. Rec. trav. chim. 40: 354. 1921.
5. BRIGL, P. Z. physiol. Chim. 116: 1. 1921.
6. BRIGL, P., GRONEMEIER, K., and SCHULZ, A. Ber. 72: 1052. 1939.
7. BRIGL, P., MÜHLSCHLEGEL, M., and SCHINLE, R. Ber. 64: 2921. 1931.
8. BRIGL, P. and SCHINLE, R. Ber. 65: 1890. 1932.
9. FRIED, J. and WALZ, DORIS E. J. Am. Chem. Soc. 71: 140. 1949.
10. GEORG, A. Helv. Chim. Acta, 15: 924. 1932.
11. HAMMETT, L. P. Physical organic chemistry. McGraw-Hill Book Co. Inc., New York. 1940.
12. HAWORTH, W. N., HIRST, E. L., and TEECE, ETHEL G. J. Chem. Soc. 1405. 1930.
13. HICKINBOTTOM, W. J. J. Chem. Soc. 3140. 1928.
14. HICKINBOTTOM, W. J. J. Chem. Soc. 1676. 1929.
15. HUDSON, C. S. J. Am. Chem. Soc. 31: 66. 1909.
16. HUDSON, C. S. *In* Advances in carbohydrate chemistry. Vol. 3. *Edited by* W. W. Pigman and M. L. Wolfrom. Academic Press Inc., New York. 1948.
17. HUDSON, C. S. and DALE, J. K. J. Am. Chem. Soc. 37: 1264. 1915.
18. HURD, C. D. and HOLYSZ, R. P. J. Am. Chem. Soc. 72: 2005. 1950.
19. ISBELL, H. S. Ann. Rev. Biochem. 9: 70. 1940.
20. JACKSON, E. L. and HUDSON, C. S. J. Am. Chem. Soc. 59: 994. 1937.
21. KUNZ, A. and HUDSON, C. S. J. Am. Chem. Soc. 48: 1982. 1926.
22. LUCAS, H. J., MITCHELL, F. W., JR. and GARNER, H. K. J. Am. Chem. Soc. 72: 2138. 1950.
23. MÜLLER, S., MÓRICZ, MARIA, and VERNER, G. Ber. 72: 745. 1939.
24. NESS, R. K., FLETCHER, H. G., JR. and HUDSON, C. S. J. Am. Chem. Soc. 73: 959. 1951.
25. PACSU, E. and WILSON, E. J., JR. J. Am. Chem. Soc. 61: 1450. 1939.
26. PACSU, E. and WILSON, E. J., JR. J. Am. Chem. Soc. 61: 1930. 1939.
27. PIGMAN, W. W. and GOEPP, R. M., JR. Chemistry of the carbohydrates. Academic Press Inc., New York. 1948.
28. RICHTMYER, N. K., CARR, C. J., and HUDSON, C. S. J. Am. Chem. Soc. 65: 1477. 1943.
29. RICHTMYER, N. K. and HUDSON, C. S. J. Am. Chem. Soc. 65: 64. 1943.
30. SCHLUBACH, H. H. Ber. 59: 840. 1926.
31. SCHLUBACH, H. H., STADLER, P., and WOLF, IRENE. Ber. 61: 287. 1928.
32. SCHNEIDER, W., SEPP, J., and STIEHLER, O. Ber. 51: 220. 1918.
33. SUNBERG, O. E. and ROYER, G. L. Ind. Eng. Chem., Anal. Ed. 18: 719. 1946.
34. WHELAND, G. W. The theory of resonance and its application to organic chemistry. John Wiley and Sons., Inc., New York. 1944.
35. WINSTEIN, S. and BUCKLES, R. E. J. Am. Chem. Soc. 64: 2780, 2787. 1942.
36. WOLFROM, M. L. and THOMPSON, A. J. Am. Chem. Soc. 56: 880, 1804. 1934.

PHOTOLYSIS OF 2, 2′, 4, 4′-TETRADEUTERODIETHYL KETONE[1]

By M. H. J. Wijnen[2] and E. W. R. Steacie

Abstract

The photolysis of $CH_3CD_2COCD_2CH_3$ has been studied over a temperature range from 25°C. to 365°C. The results confirm several features of the mechanism, previously proposed for the photolysis of diethyl ketone. It is concluded that disproportionation of ethyl radicals occurs by a "head to tail" mechanism. As activation energy for the reaction

$$CH_3CD_2 + CH_3CD_2COCD_2CH_3 = CH_3CD_3 + CH_3CDCOCD_2CH_3 \quad (4)$$

a value $E_4 = 8.7$ kcal. was found. As activation energy for Reaction (5)

$$CH_3CD_2 + CH_3CD_2COCD_2CH_3 = CH_3CD_2H + CH_2CD_2COCD_2CH_3 \quad (5)$$

a value of $E_5 = 11.7$ kcal. was found. An activation energy of $\backsim$ 17 kcal. is estimated for the thermal decomposition of the pentanonyl radical.

Introduction

As a result of previous investigations at Rochester and at Ottawa the following reactions were given (2) to explain the mechanism of the photolysis of diethyl ketone.

$C_2H_5COC_2H_5 + h\nu$	$= 2C_2H_5 + CO$	I_a	(1)
$C_2H_5 + C_2H_5$	$= C_4H_{10}$	$k_2(C_2H_5)^2$	(2)
$C_2H_5 + C_2H_5$	$= C_2H_6 + C_2H_4$	$k_3(C_2H_5)^2$	(3)
$C_2H_5 + C_2H_5COC_2H_5$	$= C_2H_6 + C_2H_4COC_2H_5$	$k_4(C_2H_5)(C_2H_5COC_2H_5)$	(4)
$C_2H_4COC_2H_5$	$= C_2H_4 + CO + C_2H_5$	$k_5(C_2H_4COC_2H_5)$.	(5)

To obtain further information about this mechanism the photolysis of $CH_3CD_2COCD_2CH_3$ has been studied. Since this compound will produce CH_3CD_2 radicals it is at the same time a promising source of ethyl radicals for the study of hydrogen-abstraction reactions of the type $C_2H_3D_2 + RH = C_2H_4D_2 + R$, and work along these lines is in progress.

Experimental

The apparatus was the same as that used in the photolysis of ordinary diethyl ketone (2). One Ward still was replaced by a pump-down liquid nitrogen trap which allowed the CO fraction to be pumped off at about −220°C. The C_2 fraction was pumped off at −175°C. and divided into two parts. From one part the ethylene was removed by absorption on a mercuric acetate bead in a Blacet–Leighton apparatus (1). The residual ethane was analyzed by means of a mass spectrometer. The other part was analyzed directly by the mass spectrometer. The spectrum of peak heights of the ethane fraction obtained from the first part was subtracted from the total C_2 spectrum and so the

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ethylene spectrum obtained. The C_4 fraction was pumped off at -115°C. Owing to the complicated cracking-pattern of butane, no results were obtained for the isotopic composition of this fraction. As light source a Hanovia 500 w. type A medium-pressure quartz lamp was used.

The diethyl ketone was prepared by Dr. L. C. Leitch and Mr. A. T. Morse of this laboratory. Mass spectrometer analysis showed the following composition: $C_4D_3H_7CO$, 4.2%; $C_4D_4H_6CO$, 91.8%; $C_4D_5H_5CO$, 2.47%; and $C_4D_6H_4CO$, 1.51%. The same analysis showed that $92 \pm 1\%$ of the ethyl radicals (produced by cracking of $C_2D_2H_3COC_2D_2H_3$) contained two deuterium atoms. Considering the method of preparation of the compound (exchange of light ketone with heavy water) it has been assumed that these two deuterium atoms are present predominantly on the α-carbon atoms. The amounts of diethyl ketones other than $CH_3CD_2COCD_2CH_3$ are, in general, too small to affect the results significantly and may be neglected in the subsequent discussion.

Results

In Table I are given the results of experiments done at various temperatures and diethyl ketone pressures. The initial intensity of the light source in several experiments was cut down by the use of neutral density filters. This is indicated in Column 2. The pressure of diethyl ketone (Column 5) has been given in mm. at °C., converted to this temperature from the pressure measured at the reaction temperature. All products are given in mm.[3] at N.T.P. Four different ethanes were found, $C_2D_4H_2$, $C_2D_3H_3$, $C_2D_2H_4$, and C_2DH_5. The amounts of $C_2D_4H_2$ produced (given in Column 10) are too small to allow accurate determination. The amounts of C_2DH_5 produced (Column 13) are, owing to overlapping of the several ethane spectra, to be considered as very inaccurate. The error for this ethane may amount to ca. 50%. It is, however, to be expected that the error will be consistent for all the determinations. Inaccurate also are the values for C_2DH_3 (last column). A much higher precision is obtainable for $C_2D_3H_3$, $C_2D_2H_4$, and $C_2D_2H_2$ and these are the most significant from the point of view of the mechanism.

To give a more simple picture of the C_2 fraction, Table II shows the components of the ethane and ethylene fraction as percentages.

Interpretation of Results

In the introduction the reaction mechanism for the photolysis of light diethyl ketone has been given. The photolysis of $CH_3CD_2COCD_2CH_3$ produces $C_2H_3D_2$ radicals which by Reaction (2) may recombine to form butane. Reactions (3), (4), and (5) become rather complex owing to the fact that here the ethyl radical contains H and D, and several isotopic reactions are thus possible in each case. We will, therefore, discuss the disproportionation reactions, the hydrogen-abstraction reactions and the thermal decomposition of the pentanonyl radicals in separate sections.

TABLE I

PRODUCTS OF THE PHOTOLYSIS OF $C_4H_6D_4CO$

Expt. No.	Intensity, relative	Temp., °C.	Time, sec.	Pressure of diethyl ketone, mm. at °C.	Products in mm.³ (N.T.P.)				Isotopic analysis					
									Ethane fraction				Ethylene fraction	
					CO	Ethane	Ethylene	C_4	$C_2D_4H_2$	$C_2D_3H_3$	$C_2D_2H_4$	C_2DH_5	$C_2D_2H_2$	C_2DH_3
6	100	24	6090	25.9	232	31.0	23.0	206	0.26	0.6	25.4	4.7	20.4	2.6
7	10	24	34530	26.8	176	24.0	19.0	151	0.35	1.3	19.3	3.1	17.6	1.4
19	10	50.5	41640	23.2	218	27.2	20.3	185	0.0	3.4	21.0	2.8	19.4	0.9
20	100	50.5	5040	25.5	261	30.8	25.4	221	0.0	1.9	26.8	2.1	22.2	3.2
16	100	101	3215	20.7	159	26.4	9.2	133	1.3	4.6	18.4	2.0	8.3	0.9
17	10	101	45180	17.5	196	41.6	15.2	155	0.4	13.3	21.0	6.9	13.3	1.9
15	10	105.5	38400	22.2	197	39.3	12.9	152	0.4	14.0	19.0	5.9	11.5	1.4
27	10	138	43200	19.4	222	70.0	21.0	145	0.0	37.1	22.5	10.3	15.3	5.7
28	10	138	56100	14.3	205	61.9	16.7	139	0.3	29.9	21.5	10.1	14.7	2.0
1	100	180	3445	19.0	176	56.0	9.5	123	2.1	23.2	18.0	12.6	8.0	1.5
2	100	180	4160	19.5	218	71.0	15.4	137	3.1	29.3	22.0	16.6	–	–
3	10	180	24720	18.8	140	70.5	14.0	62	1.6	44.7	18.8	5.5	11.8	2.2
4	100	180	1960	17.9	98	31.4	8.6	69	–	–	–	–	–	–
5	100	180	7440	6.7	151	35.0	13.0	108	1.0	14.6	16.8	2.9	–	–
10	100	236	3035	16.4	150	94	16.0	70	0.5	56.6	21.9	14.8	14.0	2.0
11	10	236	37680	16.4	202	20.4	20.0	44	1.3	132	40.2	30.6	17.1	3.0
12	10	236	45420	9.5	157	138	19.7	36	0.1	84.8	27.2	25.5	18.0	2.0
13	26.5	236	14000	16.6	167	140	17.0	46	0.6	97	30.8	28.6	14.4	2.6
44	100	250	3320	15.9	166	119	11.4	73	0.0	73.8	23.5	21.2	11.4	–
46	30	250	9675	15.9	–	146	19.0	37.0	0.0	89.7	27.4	28.4	16.6	2.4
45	10	250	34680	16.2	–	238	28.0	–	0.0	149	41.6	48.0	22.1	5.9
47	3	250	72000	16.6	–	179	20.6	–	0.0	111	31.2	37.0	16.7	3.9
14	100	273	3000	16.65	163	143	21.6	42.4	0.5	99	32.2	32.2	18.3	3.3
23	100	300	2850	15.5	163	158	38.0	29.1	0.5	93	33.8	30.7	30.2	8.6
24	100	300	2400	13.65	131	129	30.0	25.2	0.4	75.6	26.4	26.4	23.5	6.5
50	30	300	5400	16.8	–	131	36.0	–	0.0	74.5	28.3	28.3	28.3	7.7
54	10	300	14280	16.75	–	150	46.4	–	0.0	83.2	35.4	31.4	34.2	12.2
52	3	300	80220	16.5	–	324	138	–	0.0	155	87.4	81	96.5	41.5
56	100	331	2100	15.0	–	149	62.6	–	0.0	75.6	37.7	34.8	44.9	17.7
40	100	350	1980	15.3	263	264	140	25.0	0.0	119	75.5	70	97.5	42.5
25	100	365	3000	14.1	338	403	272	25.4	1.8	165	138	98	190	82
26	100	365	1260	14.9	180	169	115	15.9	0.8	70.5	55.4	42	84.4	30.1
51	30	365	2280	14.2	–	165	112	–	0.0	60.3	61.9	42.8	81.0	31.0
48	10	365	7080	15.0	–	292	232	–	0.0	92.4	115	84	162	70
53	1.1	365	57600	15.0	–	97	91	–	0.0	23.4	45.8	27.8	–	–

NOTE:—*Indicates that no determination was made, frequently because of accidental loss of the sample.*

TABLE II

ISOTOPIC COMPOSITION OF C_2 FRACTION

Expt. No.	Ethane, cc. N.T.P.	Ethylene, cc. N.T.P.	Ethane, %				Ethylene, %	
			$C_2D_4H_2$	$C_2D_3H_3$	$C_2D_2H_4$	C_2DH_5	$C_2D_2H_2$	C_2DH_3
6	0.031	0.023	0.8	2.0	82.1	15.1	89	11
7	0.024	0.019	1.4	5.5	80.3	12.8	92.5	7.5
19	0.0272	0.0203	–	12.4	77.3	10.30	99	1
20	0.0308	0.0254	–	6.3	86.9	6.8	98	2
16	0.0264	0.0092	4.7	17.9	69.8	7.6	90.5	9.5
17	0.0416	0.0152	0.9	32.0	50.6	16.5	87.5	12.5
15	0.0393	0.0129	1.0	35.6	48.4	15.2	89.6	10.4
27	0.070	0.021	–	53.0	32.2	14.8	–	–
28	0.0619	0.0167	0.5	48.4	34.8	16.3	–	–
1	0.056	0.0095	3.7	41.5	32.1	22.6	84.5	15.5
2	0.071	0.0154	4.4	41.2	31.0	23.4	–	–
3	0.0705	0.0140	2.2	63.4	26.6	7.8	84.2	15.8
4	0.0314	0.0086	–	–	–	–	–	–
5	0.035	0.013	3.1	41.8	46.9	8.2	–	–
10	0.094	0.016	0.5	60.3	23.3	15.8	87.6	12.4
11	0.204	0.020	0.6	64.7	19.7	15.0	85.6	14.4
12	0.1375	0.0197	0.1	61.6	19.7	18.6	90	10
13	0.140	0.017	0.4	61.8	19.6	18.2	84.6	15.4
44	0.118	0.0114	0.0	62.3	19.8	17.9	100	0
46	0.146	0.019	0.0	61.7	18.8	19.5	87.5	12.5
45	0.238	0.028	0.0	62.4	17.5	20.2	79	21
47	0.179	0.0206	0.0	62.0	17.4	20.6	81.3	18.7
14	0.143	0.0216	0.3	60.4	19.6	19.6	84.6	15.4
23	0.158	0.038	0.3	58.7	21.4	19.5	79.4	20.6
24	0.129	0.030	0.3	58.7	20.5	20.5	79.6	20.4
50	0.131	0.036	0.0	56.8	21.6	21.6	78.6	21.4
54	0.150	0.0464	0.0	55.5	23.6	20.9	73.8	26.2
52	0.324	0.138	0.0	47.9	27.0	25.1	70	30
56	0.149	0.0626	0.0	51.0	25.4	23.6	71.7	28.3
40	0.264	0.140	0.0	44.9	28.6	26.5	69.6	30.4
25	0.403	0.272	0.5	40.9	34.2	24.5	69.5	30.5
26	0.169	0.115	0.5	41.8	32.8	24.9	63.5	26.5
51	0.165	0.112	0.0	36.5	37.5	26.0	72.4	27.6
48	0.292	0.232	0.0	31.6	39.5	28.9	69.7	30.3
53	0.097	0.091	0.0	23.4	45.8	27.8	–	–

NOTE:—*Indicates that no determination was made, frequently because of accidental loss of the sample.*

A. Disproportionation Reactions

$$CH_3CD_2 + CH_3CD_2 = C_2H_3D_3 + C_2H_3D \quad (3a)$$

$$CH_3CD_2 + CH_3CD_2 = C_2H_4D_2 + C_2H_2D_2. \quad (3b)$$

It has been shown that in the photolysis of diethyl ketone the disproportionation reaction is alone responsible for the ethylene produced at low temperatures. In the runs at 24°C. and 50.5°C. more than 90% of the ethylene produced consists of $C_2D_2H_2$ indicating that Reaction (3*b*) is much more important than (3*a*). If we assume that C_2DH_3 found at this temperature is produced by Reaction (3*a*) then we should expect to find at least an equivalent amount of $C_2H_3D_3$. This is not so in Experiments 6 and 19. Later it will be shown that all the $C_2H_3D_3$ found at these temperatures may be accounted for by other reactions. The small amount of C_2DH_3 formed at these temperatures must therefore be attributed to the presence of ketone with less than four D atoms and we may rule out Reaction (3*a*). If $C_2D_2H_2$ is formed by dispro-

portionation and if, as shown previously, both disproportionation and combination of ethyl radicals have very low activation energies, then $R_{C_2D_2H_2}/R_{C_4}$ should be constant. Column 5 (Table III) shows this is true for runs from 24°C. to 180°C. The increase in $C_2D_2H_2$ at 180°C. and higher temperatures may be explained by other reactions as will be shown later. For the ratio of the rate constants for disproportionation and combination we obtain $k_{3b}/k_2 \backsim 0.1$.

TABLE III

RATE CONSTANT RELATIONSHIPS

Expt. No.	Temp., °C.	$R_{C_4} \cdot 10^{12}$	$\frac{R_{C_4}^{\frac{1}{2}}}{(D)} \cdot 10^{12}$	$\frac{R_{C_2D_2H_2}}{R_{C_4}}$	$k_4/k_2^{\frac{1}{2}} \cdot 10^{14}$	$k_5/k_2^{\frac{1}{2}} \cdot 10^{13}$
6	24	9.1	3.3	0.10	0.99	0.77
7	24	1.18	1.14	0.12	0.99	0.31
19	50.5	1.19	1.33	0.10	2.45	0.18
20	50.5	11.8	3.79	0.10	3.28	0.80
16	101	11.2	4.57	0.06	15.8	0.17
17	101	0.92	1.55	0.09	13.3	0.41
15	105.5	1.06	1.31	0.08	12.0	0.32
27	138	0.90	1.38	0.11	35.3	0.76
28	138	0.67	1.60	0.11	34.6	0.89
1	180	9.6	4.59	0.06	86.2	2.12
2	180	8.8	4.32	–	92.5	2.62
3	180	0.67	1.24	0.19	89.5	2.52
4	180	9.4	4.84	–	–	–
5	180	3.9	8.31	–	111	4.6
10	236	6.2	4.27	0.20	348	9.2
11	236	0.31	0.96	0.39	291	7.9
12	236	0.21	1.38	0.50	323	9.0
13	236	0.88	1.60	0.31	340	9.2
44	250	5.9	4.30	0.16	438	9.7
46	250	1.0	1.79	0.45	433	11.3
14	273	3.8	3.30	0.43	772	21.8
23	300	2.7	3.01	1.04	957	31.9
24	300	2.8	3.89	0.93	1170	36.6
40	350	3.4	3.40	3.9	1610	99.2
25	365	2.3	3.03	7.5	1970	162
26	365	3.4	3.49	5.3	1550	118

B. Hydrogen Abstraction Reactions

$$CH_3CD_2 + CH_3CD_2COCD_2CH_3 = C_2H_3D_3 + CH_3CDCOCD_2CH_3 \quad (4)$$

$$CH_3CD_2 + CH_3CD_2COCD_2CH_3 = C_2H_4D_2 + CH_2CD_2COCD_2CH_3. \quad (5)$$

The attack of an ethyl radical on diethyl ketone may result in the removal of a primary or of a secondary hydrogen atom, i.e. of a D-atom (Reaction (4)) or of an H-atom (Reaction 5)). Reaction (4) produces $C_2H_3D_3$ which has been identified as one of the reaction products. Following the derivations made in the photolysis of diethyl ketone we find that if Reaction (4) occurs,

$$k_4/k_2^{\frac{1}{2}} = \frac{R_{C_2D_3H_3}}{R_{C_4}} \times \frac{R_{C_4}^{\frac{1}{2}}}{(D)}. \qquad \text{I}$$

Independent of pressure or intensity used, we should find constant values for

$k_4/k_2^{\frac{1}{2}}$ at constant temperature. The results obtained for $k_4/k_2^{\frac{1}{2}}$ (Column 6, Table III) confirm Equation I. By plotting log $k_4/k_2^{\frac{1}{2}}$ against $1/T$ (Fig. 1) we obtain, assuming $E_2 = 0$ kcal., as activation energy for Reaction (4), $E_4 = 8.7$ kcal.

An equation similar to Equation I may be derived for Reaction (5). We have to consider, however, that the ethane produced by this reaction ($C_2H_4D_2$)

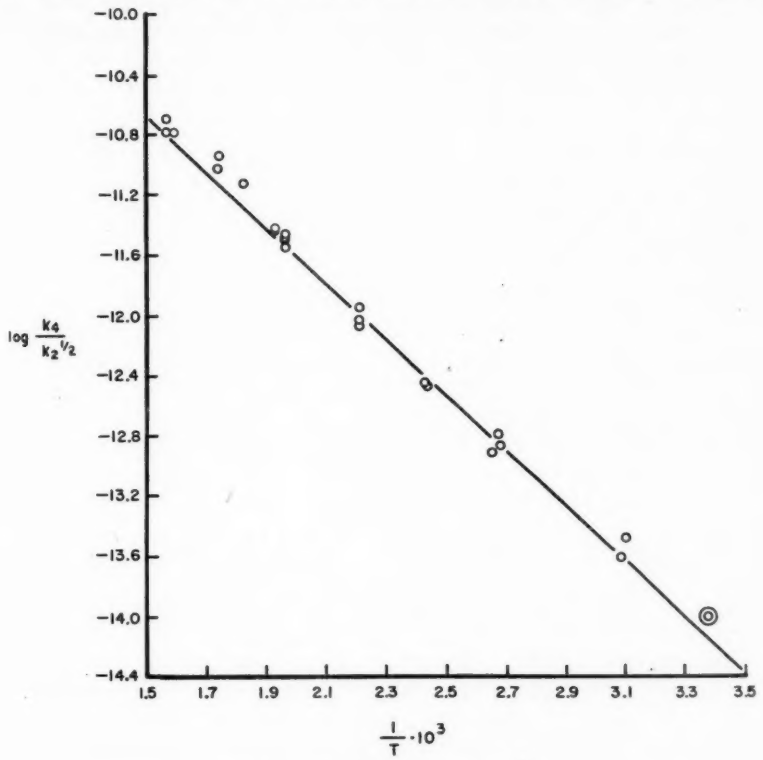

FIG. 1. *Plot of log* $k_4/k_2^{\frac{1}{2}}$ *against* $\frac{1}{T}$.

is also produced by Reaction (3*b*). We therefore have to subtract the ethane produced by disproportionation from the total amount of $C_2H_4D_2$ found. The ethane produced by disproportionation is equal to the $C_2H_2D_2$ produced. A simple subtraction of $C_2H_2D_2$ from $C_2H_4D_2$ would, however, not do since at high temperatures, as is already indicated and will be shown in the next section, $C_2H_2D_2$ is also produced by other reactions. In Section *A*, however, it was shown that

$$k_2/k_4 = \frac{R_{C_2D_2H_2}}{R_{C_4}} \sim 0.1.$$

To obtain the amount of $C_2H_4D_2$ produced by Reaction (5) only, we have, therefore, subtracted 1/10 of the amount of C_4 produced from the total amount of $C_2H_4D_2$. We thus obtain Equation II

$$k_5/k_2^{\frac{1}{2}} = \frac{R_{C_2H_2D_2} - 1/10\ R_{C_4}}{R_{C_4}} \times \frac{R_{C_4}^{\frac{1}{2}}}{(D)}. \qquad \text{II}$$

The values of $k_5/k_2^{\frac{1}{2}}$ obtained are given in Column 7 (Table III). It will be seen that the equation holds very approximately, which is not surprising in

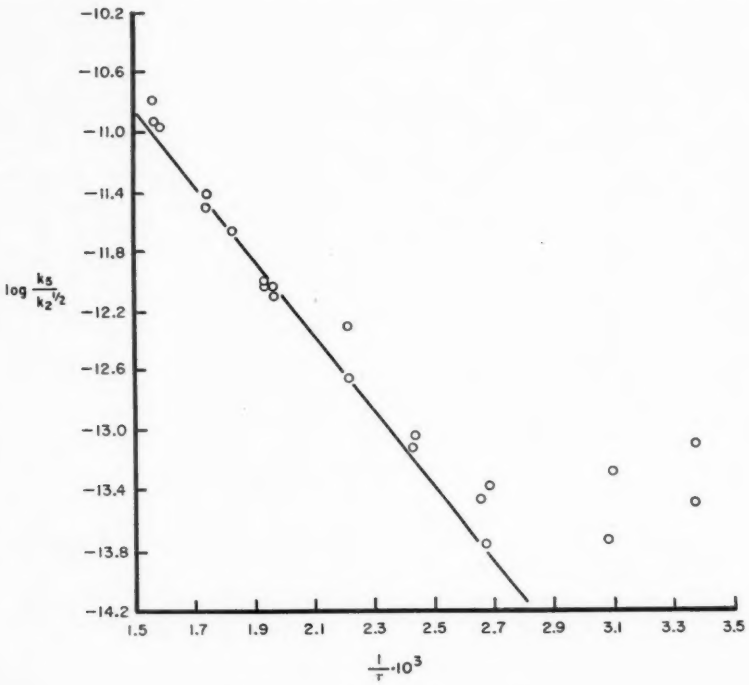

FIG. 2. *Plot of log* $k_5/k_2^{\frac{1}{2}}$ *against* $\frac{1}{T}$.

view of the approximate nature of the method of calculation. If log $k_5/k_2^{\frac{1}{2}}$ is plotted against $1/T$ (Fig. 2) we obtain as activation energy for Reaction (5) (assuming $E_2 = 0$) $E_5 = 11.7$ kcal. The amount of $C_2D_2H_4$ produced by Reaction 5 is at low temperatures very small, and hence there is a large scatter at low temperatures in Fig. 2. At higher temperatures, however, the plot is reasonably linear. The value of E_5 thus obtained is certainly much less precise than that of E_4 where the calculation is straightforward. There seems to be no doubt, however, that E_5 is considerably greater than E_4. This point will be discussed later.

C. Thermal Decomposition of the Pentanonyl Radical

$$CH_3CDCOCD_2CH_3 = C_2H_3D + CO + CD_2CH_3 \quad (6)$$

$$CH_2CD_2COCD_2CH_3 = C_2H_2D_2 + CO + CD_2CH_3 \quad (7)$$

The increase of $R_{C_2H_4}/R_{C_4H_{10}}$ at high temperatures and/or low intensity has in the photolysis of diethyl ketone been explained by a thermal decomposition of the $C_2H_4COC_2H_5$ radical. The present investigation throws considerably more light on this decomposition. Owing to various experimental complications, however, the results discussed in this section are open to some doubt, and the conclusions drawn are, therefore, somewhat speculative.

The pentanonyl radical is formed by Reactions (4) and (5) above. The amount of ethylene produced by thermal decomposition of the radicals $CH_3CDCOCD_2CH_3$ and $CH_2CD_2COCD_2CH_3$ can thus never exceed the amount of $C_2D_3H_3$ and $C_2D_4H_2$ produced. Table I shows that C_2DH_3 is always smaller than $C_2D_3H_3$. At high temperatures, however, the amount of $C_2D_2H_2$ exceeds the amount of $C_2D_2H_4$, indicating that $C_2D_2H_2$ is not formed solely by Reaction (5). (The amount of $C_2D_2H_2$ formed by disproportionation is negligible at these temperatures.)

It may be seen from Table I that at high temperatures (except for Runs 48 and 53 at low intensities) the amount of $C_2D_3H_3$ formed is always larger than $C_2D_2H_4$. It would, therefore, be expected that more C_2H_3D would be formed than $C_2H_2D_2$, but the reverse is the case. It is difficult to see why this should be true, and the situation is too complex to permit definite conclusions. Possible explanations are:

(*a*) A higher activation energy for the decomposition of $CH_3CDCOCD_2CH_3$ than for $CH_2CD_2COCD_2CH_3$.

(*b*) A reaction of the type
$$CH_3CDCOCD_2CH_3 + CH_3CD_2COCD_2CH_3 = CH_3CHDCOCD_2CH_3 + CH_2CD_2COCD_2CH_3.$$
The abstraction of a primary rather than a secondary hydrogen in such a reaction might be favored on steric grounds.

(*c*) An internal exchange, or isomerization, of the pentanonyl radical
$$CH_3CDCOCD_2CH_3 = CH_3CHDCOCD_2CH_2.$$

(*d*) Rearrangement of the radical on decomposing
$$CH_3CDCOCD_2CH_3 = CH_2CD_2 + CO + CH_3CHD.$$

On the above basis we may write for the decomposition of the radical, in addition to (6) and (7) also

$$CH_3CDCOCD_2CH_3 = C_2H_2D_2 + CO + C_2H_4D. \quad (8)$$

If Reaction (8) occurs it will lead to an increase in C_2H_5D at high temperatures as is found.

If the mechanism is assumed to include Reactions (1) to (8), together with the obvious recombination reactions

TABLE IV

Expt. No.	Temp., °C.	Relative intensity	$R_{C_4}.10^{12}$	$R_{C_2D_2H_2}.10^{12}$	$R_{C_2DH_3}.10^{12}$	$\left(\frac{R_{C_2D_2H_2}-1/10\,R_{C_4}}{(D)}\right).10^8$	$\frac{R_{C_2DH_3}}{(D)}.10^8$
27	138	10	0.904	0.095	0.035	0.67	5.1
28	138	10	0.665	0.071	0.0096	0.89	1.9
1	180	100	9.57	0.62	0.117	–	17.4
3	180	10	0.675	0.128	0.024	9.2	3.6
10	236	100	6.16	1.24	0.178	107	30.7
13	236	26.5	0.88	0.277	0.050	31.9	8.5
11	236	10	0.310	0.122	0.0206	15.7	3.5
12	236	10	0.214	0.106	0.0118	25.2	3.5
44	250	100	5.87	0.92	–	59	–
46	250	30	1.03	0.463	0.067	64	11.9
45	250	10	(0.214)	0.175	0.045	27	7.9
47	250	3	(0.026)	0.062	0.0145	10.1	2.5
14	273	100	3.80	1.64	0.296	214	50
23	300	100	2.74	2.84	0.81	470	148
24	300	100	2.82	2.62	0.72	485	149
50	300	30	(0.378)	1.41	0.383	230	64.4
54	300	10	(0.077)	0.645	0.230	109	39
52	300	3	(0.0086)	0.322	0.139	55	24
56	331	100	(2.72)	5.27	2.27	930	427
40	350	100	3.40	13.3	5.79	2390	1070
25	365	100	2.28	17.0	7.35	3360	1475
26	365	100	3.40	18.1	6.42	3370	1220
51	365	30	(0.83)	9.56	3.66	1880	727
48	365	10	(0.18)	6.15	2.66	1155	500

$$CH_3CD_2 + CH_3CDCOCD_2CH_3 = X \quad (9)$$

and

$$CH_3CD_2 + CH_2CD_2COCD_2CH_3 = X, \quad (10)$$

then a steady state treatment leads to

$$\frac{R_{C_2DH_3}}{[D]} = \frac{k_6k_4(CH_3CD_2)}{k_7 + k_8 + k_9\ (CH_3CD_2)} \quad \text{III}$$

The values obtained for $R_{C_2DH_3}/(D)$ are given in Table IV for experiments at higher temperatures where the decomposition of the pentanonyl radical becomes important. Except at low temperatures where the amount of C_2DH_3 is very small the results are consistent, and at a given temperature $R_{C_2DH_3}/D$ decreases with decreasing temperature as required by Equation III.

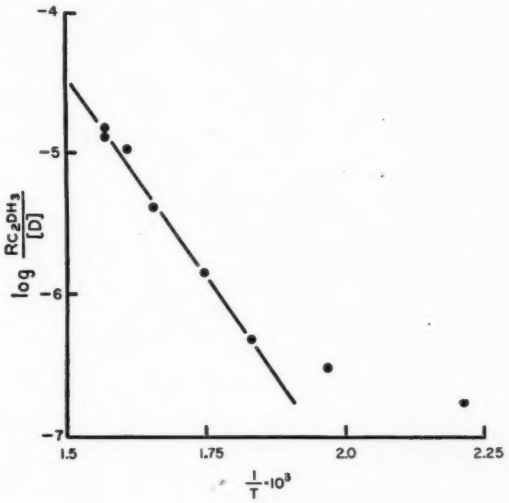

FIG. 3. *Plot of log* $\frac{R_{C_2DH_3}}{[D]}$ *against* $\frac{1}{T}$.

At the highest intensities it may be assumed that $k_7 + k_8$ will be negligible compared with $k_9(CH_3CD_2)$. Equation III then becomes

$$\frac{R_{C_2DH_3}}{[D]} \backsim \frac{k_6k_4}{k_9}. \quad \text{IV}$$

This is not a very good approximation, but is about the only thing that can be done. In Fig. 3 $\log \frac{R_{C_2DH_3}}{(D)}$ is plotted against $1/T$ for experiments at the highest intensity. The activation energy thus obtained is

$$E_6 + E_4 - E_9 = 25.7 \text{ kcal.}$$

Taking $E_4 = 8.7$ kcal. as found above, and assuming $E_9 = 0$ as usual, this gives $E_6 \sim 17$ kcal., which seems a reasonable value.

Discussion

The most important conclusion from the present work concerns the mechanism of disproportionation. There are two plausible possibilities for this reaction, a "head to tail" mechanism

$$\boxed{CH_3.CH_2-} + H.CH_2.CH_2- = C_2H_6 + -CH_2.CH_2- \qquad (A)$$

leading to the formation of the bimethylene radical, and a "head to head" mechanism

$$\boxed{CH_3.CH_2-} + H\overset{|}{C}H.CH_3 = C_2H_6 + \diagup\!\!\diagdown CH.CH_3 \qquad (B)$$

leading to the ethylidene radical. If the biradicals thus formed had an appreciable existence the reaction would be similar to an ordinary hydrogen-abstraction reaction, and would be expected to have an activation energy of the order of 7-10 kcal. If, however, the biradicals isomerize to ethylene simultaneously with the primary step the disproportionation reaction will be strongly exothermic, and might be expected to have an activation energy of approximately zero, as found. It would appear much easier for ethylene to be formed directly in (*A*) than in (*B*) which involves a hydrogen transfer. Hence the fact that the activation energy is approximately zero strongly favors mechanism (*A*). The isotopic composition of the ethane formed by disproportionation in the present investigation gives a direct proof of the "head to tail" mechanism.

For the ratio of the rate constants of the disproportionation and recombination reactions the value obtained is $k_3/k_2 \backsim 0.1$, in agreement with the value obtained with light diethyl ketone (2).

For the hydrogen-abstraction reactions the activation energies found are $E_4 = 8.7$ kcal., $E_5 = 11.7$ kcal. The results thus indicate a considerably lower activation energy for the abstraction of a secondary hydrogen as compared with a primary hydrogen. It should be noticed that in one case a D– and in the other an H-atom is abstracted. Isotopic effects will influence the relative magnitudes of the activation energies obtained and will probably even enlarge this difference.

In the reaction $C_2H_5 + C_2H_5COC_2H_5 = C_2H_6 + C_2H_4COC_2H_5$ there is no means of deciding where the H-atom has been abstracted. The relatively low activation energy (E = 7.4 kcal. (2)) however indicates that the activation energy measured is the one for abstraction of a secondary hydrogen.

The fact that the activation energy for the abstraction of a secondary hydrogen is considerably lower than that for a primary hydrogen is in line with the results for methyl radical reactions (4). It is also in line with chemical evidence that the "activity" of hydrogen atoms is enhanced by the presence of a neighboring carbonyl group.

The fact that the activation energy of Reaction (4) is higher than the similar

one in $C_2H_5 + C_2H_5COC_2H_5$ agrees with results obtained in the photolysis of acetone. Dr. T. G. Majury (3) of this laboratory obtained $E = 9.6$ kcal. for the abstraction of an H atom from acetone by a CH_3 radical, and $E = 11.4$ for the abstraction of a D atom from heavy acetone by a CD_3 radical. Accepting for the collision diameters of diethyl ketone and the $C_2H_3D_2$ radical, 6 and 4×10^{-8} cm. respectively, the following values are obtained for the steric factor ratios $P_4/P_2^{\frac{1}{2}} \backsim 5.10^{-4}$ and $P_5/P_2^{\frac{1}{2}} \backsim 3.10^{-3}$. The corresponding ratio was found to be $\sim 10^{-3}$ in the photolysis of light diethyl ketone (2).

Acknowledgment

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References

1. BLACET, F. E., MACDONALD, G. D., and LEIGHTON, P. A. Ind. Eng. Chem., Anal. Ed. 5: 272. 1933.
2. KUTSCHKE, K. O., WIJNEN, M. H. J., and STEACIE, E. W. R. J. Am. Chem. Soc. In press.
3. MAJURY, T. G. and STEACIE, E. W. R. J. Chem. Phys. In press.
4. TROTMAN-DICKENSON, A. F. and STEACIE, E. W. R. J. Chem. Phys. 19: 329. 1951.

AN EVALUATION OF OSMOTIC PRESSURE MEASUREMENTS USING POLYVINYL ACETATE SYSTEMS[1]

By L. A. McLeod[2] and R. McIntosh[3]

Abstract

Measurements of osmotic pressures of polyvinyl acetate solutions in various solvents have been made. These pressures are precise to ±0.005 cm. of solvent and can be repeated on successive samples to 0.02 cm. In five solvents, plots of reduced osmotic pressure (π/C_2) against concentration show distinct upward curvature which differs significantly from theoretical equations derived by Flory and by Huggins. The evidence suggests that further refinement of these theoretical equations is required to bring them into exact agreement with experimental data.

Introduction

Since polymer solutions were first found to depart from the behavior predicted by van't Hoff's law, attempts have been made to rationalize their observed osmotic behavior. The most satisfactory description of the behavior of such solutions is the equation proposed by Huggins (5) and derived independently by Flory (3) and Miller (7). They suggested a quadratic dependence of reduced osmotic pressure on concentration, usually used in the form

$$\frac{\pi}{C_2} - \frac{RTd_1}{3M_1d_2^3}C_2^2 = \frac{RT}{M_2} + \frac{RTd_1}{M_1d_2^2}(1/2 - \mu_1)C_2$$

where π is osmotic pressure, C_2 is solute concentration, d_1 and d_2 are densities of solvent and solute respectively, M_1 and M_2 are molecular weights of solvent and solute respectively, μ_1 is a constant for a particular polymer–solvent system, R is the molar gas constant, and T is the absolute temperature. This equation has served to express the osmotic pressure data reported for many polymer solutions.

Attempts to determine accurate osmotic pressure values led Sirianni, Wise, and McIntosh (9) to the conclusion that metal osmometers of the usual design were inadequate. These authors showed that the use of a membrane as a gasket may lead to erroneous results; they also observed a difference in osmotic behavior for different types of membrane but their evidence was at variance with the results published by Robertson, McIntosh, and Grummitt (8). These discrepancies were of such a fundamental nature that it was decided to continue the study of the measurement of osmotic pressure *per se*. The purpose of the following investigation has therefore been to develop a technique of osmotic measurements of sufficient precision to show whether or not the osmotic pres-

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sure is influenced by the nature of the membrane; whether the intercepts of the π/C_2 vs. C_2 curves are identical for different solvents; whether van't Hoff's law describes the effect of temperature on the limiting value of π/C_2; and other such basic questions. The effect of solvent type is discussed in the present report.

Two points of interest arise in the analysis of experimental osmotic data. The first is the form of the π/C_2 vs. C_2 curve. The slope of this line and the presence or absence of curvature are directly related to the solvent–polymer interaction forces. Reference has already been made to the theoretical equation derived by Flory, Huggins, and Miller to relate π/C_2 to C_2. By modifying his original assumptions, Flory (4) has recently deduced another equation for osmotic behavior, which he states in the form

$$\frac{\pi}{C_2} = \frac{RT}{M_2}\left[1 + \Gamma C_2 + \tfrac{5}{8}\,\Gamma^2 C_2^2\right]$$

where Γ is a constant and the other symbols are the same as in the first equation. The ability of these equations to fit experimental osmotic data should be a reflection of the validity of their underlying assumptions.

The second point of interest in the analysis of the data is the value of $(\pi/C_2)_{C_2=0}$ since this leads directly to the molecular weight of the solute. Extrapolation of the π/C_2 vs. C_2 curve to this intercept is much more difficult if curvature exists. Extrapolation may be done visually but with this method it is difficult to be completely objective; an empirical equation may be derived for the range in which data are available and used to calculate $(\pi/C_2)_{C_2=0}$. Bawn, Freeman, and Kamaliddin (1) have outlined a convenient procedure for this treatment; extrapolation of the curve would be most reliable if the experimental data could be fitted to a theoretical equation.

The data obtained in the present investigation are considered sufficiently dependable to assess the merits of the two theoretical equations and to compare the various ways of obtaining the extrapolated intercept.

Experimental

The Osmometer

The osmometer is only a minor adaptation of that described by Sirianni, Wise, and McIntosh (9). Their experience showed the need for a ridge surrounding the cell to enable the membrane to be fully submerged in solvent during assembly. Their difficulties attributed to corrosion of the brass cell have been largely eliminated by gold-plating the cell. Other changes have also been incorporated into the design:

1. Since cold flow of the "Composition" lead foil gave rise to leakage after about 10 days of use, this foil was replaced by a Teflon gasket which has given excellent service.

2. A more convenient method of controlling evaporation of the cell contents was to connect the two measuring capillaries together. This did not improve the drop of liquid level reported by Sirianni, Wise, and McIntosh but was a simpler method.
3. The Fernico bushings used to seal the measuring capillaries to the brass cell were found to rust in water and were replaced by copper–nonex seals.
4. It has not been possible to eliminate the trouble caused by air bubbles although the gold-plating of the cell has given a marked improvement in behavior. In spite of very careful oscillation of the liquid levels in the cell, coupled with a rocking motion of the cell itself and a slight (2-3°) warming of the cell contents, bubbles still appear occasionally and for no apparent reason. Their presence is usually manifested by a rise in levels in both capillaries.

With these relatively minor changes, the cell design described by Sirianni, Wise, and McIntosh (9) has been accepted as adequate. Another cell of the same design has been made of stainless steel (S.A.E. No. 303) but its chief advantage over the gold-plated cell lies in its resistance to the action of mercury.

Membranes

Preparation of a semipermeable membrane still constitutes a serious problem for the systems involved in this work. It was finally found necessary to fractionate the polymer to the extent described below in order to eliminate solute leakage. Dried "Cellophane" (No. 600 P.T. not waterproofed, obtained through the courtesy of Canadian Industries Ltd.), has been used throughout the present investigation but care is necessary to select sheets which are free from air bubbles. The membranes were swollen in 5% sodium hydroxide according to the procedure previously outlined (9) but the various times of swelling and accommodation to solvent were not found to be critical and the results did not appear to depend on the amount of sodium hydroxide left in the membrane. As in the case of the earlier investigations (8, 9), these caustic-swollen "Cellophane" membranes showed no evidence of adsorption of solute.

A very rigorous test for semipermeability was used. Membranes intended for use with Gelva V360 (molecular weight 300,000) were tested overnight with a 3% acetone solution of Gelva V15 (molecular weight 45,000). The contents of the solvent side were then poured into an equal volume of water. Any cloudiness caused the membrane to be rejected.

Materials

Commercial polyvinyl acetate (provided through the courtesy of Shawinigan Chemicals Ltd.) was used. A partial fractionation procedure was used in order to remove the very small and very large polymers. A 5% solution of the crude polymer in acetone was filtered through a sintered glass funnel. The polymer was precipitated from the filtrate by slow addition of water to the solution until only a faint cloudiness resulted from further addition of water. The

polymer was allowed to settle out, decanted, redissolved in acetone, and reprecipitated in the same manner. After redissolving the polymer in acetone, the final precipitation was effected by slow addition of the acetone solution to a large excess of water. This produced a very spongy type of polymer which, after partial drying, was sliced thin and dried *in vacuo* at room temperature.

Reagent grade solvents stored over Drierite were used without further treatment.

Preparation of Solutions

Concentrations were measured either from known weights of solute and solvent or by evaporation to constant weight of an aliquot of the solution. Results from the two methods agreed within 1% when used on the same solutions. It was thus possible to use the second method with confidence to measure concentrations of solutions obtained by dilution from a stock solution. Concentrations are probably reliable to 0.2% in the upper range and 0.5% at the lower range.

Temperature Control

Measurements were made at 30°C. in a water thermostat controlled to ±0.002°C. (i.e. no fluctuation was observed on a Beckmann thermometer). This control can be achieved by using an American Instrument Co. Metastatic thermoregulator and any convenient electronic relay, provided the regulator is vibrated periodically to prevent sticking at the mercury surface.

Measurements

For greatest precision the cell was used as a static osmometer. Equilibrium pressures approached from above and below were averaged. The two readings usually agreed to ±0.005 cm. of the mean. Equilibrium was attained in about five hours and was followed overnight to detect any drift due to solute leakage. If a drift occurred overnight, the reading was rejected and the membrane was tested again with Gelva V15.

Periodically the cell was rinsed with solvent and a measurement taken of the "cell zero". If repeated washing with solvent did not reduce the "cell zero" to less than 0.01 cm., the readings were rejected and the cell was tested for failure of the gasket since a large "cell zero" can usually be attributed to leakage at this location (1, 9).

Results

Three polymer samples were used in this work and are designated Gelvas V360, V360A, and V 360B respectively. They are the result of three similar preparations from the same commercial sample. Gelvas V360 and V360B appeared to have the same molecular weight and so are treated as identical samples; Gelva V360A showed a slightly lower molecular weight and was dealt with separately. The two osmometers (the gold-plated brass cell and the stainless steel cell) were used interchangeably except in the case of Gelva V360A in benzene where an unaccountable difference in slope but not in intercept was

TABLE I

μ_1 VALUES FOR POLYVINYL ACETATE IN VARIOUS SOLVENTS

Solvent	McLeod and McIntosh	Sirianni, Wise, and McIntosh(9)	Masson and Melville(6)	Browning and Ferry(2)
Acetone	0.439 (30°)	0.439 (30°)		0.437 (25°)
Methyl ethyl ketone	0.435 (30°)			0.429-0.44 (25°)
Benzene	0.427-0.430 (30°)	0.427 (30°)	0.44 (20°)	
Chloroform	0.338-0.345 (30°)	0.377 (30°)		
Ethyl acetate	0.412-0.423 (30°)		0.415 (20°)	

observed. A total of five solvents was used: acetone, benzene, methyl ethyl ketone, chloroform, and ethyl acetate.

The data were plotted on graphs of π/C_2 vs. C_2 and in every case upward curvature was evident (Fig. 1). However, all curves could be readily extrapolated to a common intercept which was 0.735 for Gelvas V360 and V360B and 0.875 for Gelva V360A.

The data were then plotted in the form $\left(\frac{\pi}{C_2} - \frac{RTd_1}{3M_1d_2^3}C_2^2\right)$ vs. C_2. According to the Flory–Huggins theory this treatment should yield a straight line but it failed to remove all the curvature in the present data. The μ_1 values

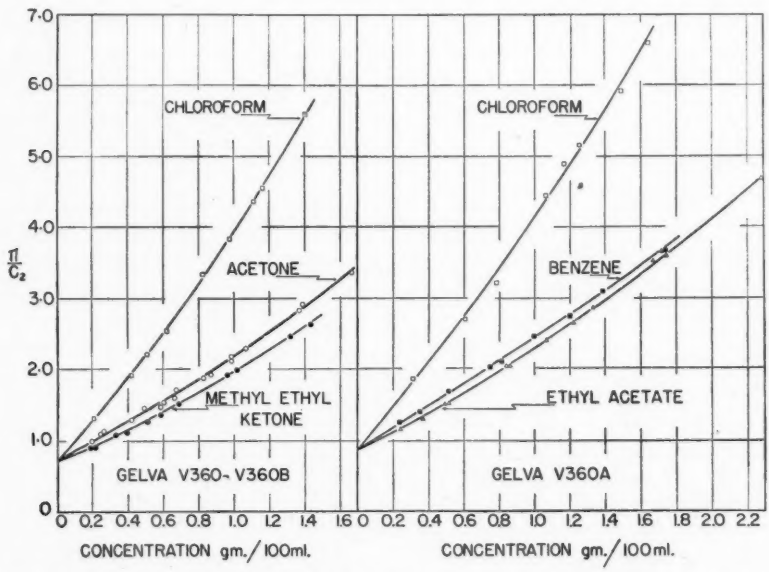

FIG. 1. *Representative graphs of reduced osmotic pressure as a function of concentration.*

obtained from these curves are listed in Table I and are in excellent agreement with the results of Sirianni, Wise, and McIntosh (9), Masson and Melville (6), and Browning and Ferry (2) for comparable systems.

This residual curvature in the Flory–Huggins plot (reported also by Masson and Melville(6)) suggests that the Flory–Huggins equation does not adequately describe the concentration dependence of π/C_2. An attempt was therefore made to define the curvature of the π/C_2 vs. C_2 plot in the following manner. The experimental data were fitted by the method of least squares to three quadratic equations having the general form $\pi/C_2 = a_0 + a_1C_2 + a_2C_2^2$. In the first case, (the Flory–Huggins equation) $a_0 = \frac{RT}{M_2}$, $a_1 = \frac{RTd_1}{M_1d_2^2}$ $(\frac{1}{2} - \mu_1)$, and $a_2 = \frac{RTd_1}{3M_1d_2^3}$. The symbols have the same significance as outlined above.

TABLE II

DERIVED CONSTANTS FOR THEORETICAL PARABOLAS

Solvent	Huggins		Flory		Least squares		
	a_0	μ_1	a_0	Γ	a_0	a_1	a_2
Gelva V360-V360B							
Acetone	0.644	0.439	0.891	0.890	0.776	1.107	0.276
Methyl ethyl ketone	0.615	0.435	0.789	0.917	0.684	1.065	0.206
Benzene	0.641	0.428	0.857	0.890	0.774	1.011	0.298
Chloroform	0.441	0.345	1.092	1.363	0.742	2.524	0.651
Ethyl acetate	0.659	0.423	0.881	0.854	0.841	0.866	0.344
Gelva V360A							
Benzene—brass	0.748	0.427	1.012	0.785	0.888	1.112	0.236
Benzene—steel	0.971	0.430	1.137	0.735	1.043	1.144	0.216
Chloroform	0.518	0.338	1.456	1.058	0.921	2.689	0.504
Ethyl acetate	0.685	0.413	1.055	0.758	0.785	1.359	0.155

Values of d_1 were obtained from Reference (10). For the Flory equation, $a_0 = \frac{RT}{M_2}$, $a_1 = \frac{RT}{M_2}\Gamma$, and $a_2 = \frac{5}{8}\frac{RT}{M_2}\Gamma^2$ where Γ is a constant. In the third case, a_0, a_1, and a_2 are not assigned any predetermined relationship. The calculated constants for these three equations are listed in Table II and the differences between the experimental points and the calculated values appear in Table III.

A representative set of data from Table II is shown graphically in Fig. 2. On this graph are plotted the experimentally determined values of π/C_2 vs. C_2 for Gelva V360 in chloroform and in ethyl acetate, together with the best fitting curves for the three equations calculated from these experimental data. It is seen that the parabola represented by the Flory–Huggins equation does not have enough curvature to fit the experimental points whereas the curvature required by the new Flory equation is too great to fit the experimental facts. The parabola whose constants are unrestricted by any arbitrary relationship gives very good agreement with the experimental data. The same general

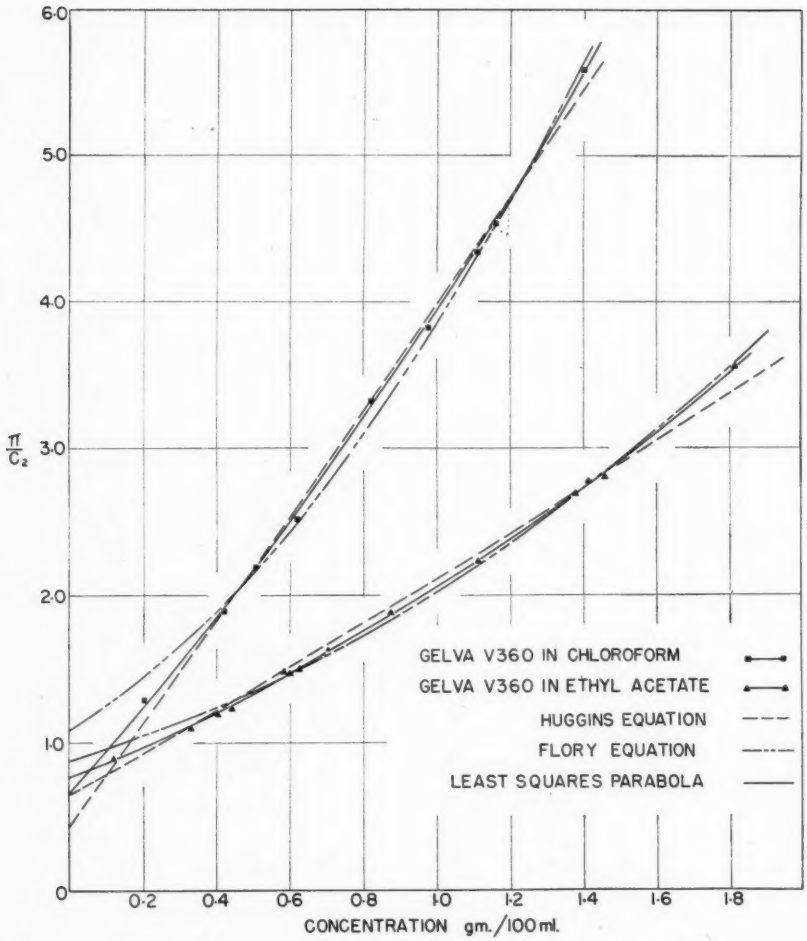

FIG. 2. *Comparison of theoretical equations derived from experimental osmotic data.*

statement applies to the intercepts on the π/C_2 axis. For both the Flory and the Flory–Huggins equations the spread in the intercepts is much greater than for the unrestricted quadratic equation.

The disagreement of the two theoretical equations with the experimental facts is illustrated much more graphically by the data of Table III. It is immediately obvious that the average deviation of the experimental points from the curve representing an unrestricted quadratic is approximately ± 0.030 cm. The average deviation from the Flory–Huggins equation and from the Flory equation is about ± 0.050 cm. This in itself suggests that the unrestricted equation is a better representation of the experimental data than is either the

TABLE III

π/C_2 VALUES FOR POLYVINYL ACETATE SYSTEMS

Gelva V360–V360B					Gelva V360–V360B				
C_2, gm./100 ml.	π/C_2 obs.	(π/C_2 obs.–π/C_2 calc.)			C_2, gm./100 ml.	π/C_2 obs.	(π/C_2 obs.–π/C_2 calc.)		
		Huggins	Flory	Least squares			Huggins	Flory	Least squares
Acetone					Benzene – brass				
1.667 B*	3.370	.055	−.069	−.018	1.860	3.658	.113	−.085	−.028
1.667 S	3.359	.044	−.080	−.029	1.451	2.899	.027	.042	.031
1.390 B	2.913	.068	.068	.065	1.382	2.820	.059	.099	.080
1.370 S	2.837	.026	.032	.026	1.320	2.593	−.069	−.010	−.035
1.067 S	2.279	−.031	.040	.008	1.162	2.308	−.102	−.008	−.043
.989 B	2.166	−.017	.059	.025	.821	1.823	−.051	.054	.018
.989 S	2.105	−.078	−.002	−.036	.751	1.690	−.076	.021	−.011
.870 B	1.920	−.071	.005	−.028	.549	1.423	−.034	.019	.004
.870 S	1.899	−.092	−.016	−.049	.504	1.330	−.059	−.019	−.029
.830 B	1.866	−.060	.013	−.019	.343	1.159	.012	−.010	.003
.830 S	1.949	.023	−.004	.064	.336	1.129	−.007	−.032	−.018
.678 B	1.701	.017	.070	.046	.278	1.084	.034	−.018	.006
.666 S	1.591	−.054	−.024	−.045	.205	.974	.033	−.057	−.020
.606 B	1.532	−.039	−.002	−.016	.176	1.006	.107	.002	.045
.587 S	1.469	−.073	−.039	−.052					
.496 B	1.452	.053	.059	.059	Mean		.056	.034	.027
.422 B	1.290	.006	−.014	−.002					
.391 B	1.354	.118	.085	.103	Chloroform – brass				
.391 S	1.111	−.125	−.158	−.140	1.405	5.583	.140	−.103	.010
.267 S	1.137	.091	.003	.046	1.165	4.548	−.023	.001	−.019
.249 S	1.101	.082	−.015	.033	1.115	4.347	−.043	.019	−.019
.200 B	1.001	.056	−.066	−.007	.980	3.839	−.064	.071	−.003
.200 S	.981	.036	−.086	−.027	.825	3.339	−.009	.156	.070
					.622	2.534	−.091	.026	−.031
Mean		.057	.044	.041	.511	2.204	−.007	.020	.002
					.425	1.906	−.022	−.048	−.027
Methyl ethyl ketone – brass					.207	1.307	.144	−.147	.015
1.438	2.634	.030	−.053	−.007					
1.321	2.445	.011	−.023	−.005	Mean		.060	.066	.022
1.020	1.994	−.008	.036	.009					
1.017	1.990	−.008	.036	.010	Ethyl acetate				
.965	1.920	−.004	.047	.016	1.819 B*	3.572	.174	−.006	.018
.691	1.511	−.031	.024	−.007	1.462 B	2.817	−.016	−.022	−.025
.588	1.374	−.026	.016	−.007	1.419 S	2.776	.009	.019	.013
.518	1.271	−.034	−.004	−.020	1.381 S	2.704	−.003	.018	.011
.400	1.120	−.025	−.025	−.023	1.353 S	2.588	−.076	−.046	−.055
.337	1.089	.029	.009	.023	1.117 B	2.248	−.052	.026	.010
.219	.918	.015	−.049	−.009	.880 S	1.891	−.051	.037	.022
.190	.911	.047	−.030	.017	.709 B	1.640	−.046	.024	.012
					.630 S	1.509	−.060	−.005	−.014
Mean		.022	.029	.013	.603 B	1.479	−.050	−.002	−.009
					.588 B	1.492	−.015	.030	−.023
					.525 S	1.467	.053	.080	.076
					.445 B	1.244	−.053	−.051	−.050
					.407 S	1.201	−.042	−.053	−.050
					.334 B	1.114	−.023	−.063	−.054
					.230 S	1.159	.172	.084	.101
					.122 B	.917	.085	−.062	−.035
					Mean		.058	.037	.034

TABLE III—*Concluded*

Gelva V360A					Gelva V360A				
C_2, gm./100 ml.	π/C_2 obs.	(π/C_2 obs.−π/C_2 calc.) Huggins	Flory	Least squares	C_2, gm./100 ml.	π/C_2 obs.	(π/C_2 obs.−π/C_2 calc.) Huggins	Flory	Least squares
Benzene – brass					Chloroform – steel				
1.818	3.691	.073	−.053	.001	1.800	7.502	.249	−.027	.108
1.551	3.170	−.003	−.012	−.011	1.644	6.587	−.067	−.155	−.117
1.517	3.131	−.014	.017	.013	1.490	5.916	−.148	−.097	−.130
1.503	3.087	−.007	.000	−.005	1.257	5.147	−.031	.145	.050
1.125	2.416	−.065	.017	−.022	1.169	4.878	.032	.229	.125
1.103	2.408	−.037	.045	.006	1.063	4.443	−.003	.203	.094
.846	2.010	−.028	.047	.012	.782	3.220	−.174	−.063	−.112
.844	1.971	−.063	.011	−.024	.609	2.696	−.055	−.075	−.050
.610	1.639	−.030	−.003	−.015	.317	1.864	.190	−.182	.040
.582	1.681	.055	.075	.066					
.444	1.378	−.036	−.063	−.050	Mean		.105	.131	.092
.422	1.426	.045	.009	.027					
.275	1.231	.073	−.029	.019	Ethyl acetate – brass				
.271	1.179	.027	−.077	−.028	2.286	4.682	.081	−.181	−.020
					1.749	3.599	−.034	−.012	−.037
Mean		.040	.033	.021	1.724	3.650	.061	.090	.061
					1.675	3.525	.024	.067	.029
Benzene – steel					1.335	2.847	−.060	.049	−.028
1.743	3.656	.046	−.104	−.037	1.237	2.753	.015	.129	.050
1.387	3.092	.049	.057	.047	1.222	2.646	−.066	.048	−.031
1.205	2.739	−.020	.037	.004	1.068	2.395	−.053	.054	−.018
1.000	2.454	.011	.097	.051	.863	2.027	−.073	.000	−.046
.815	2.089	−.074	.016	−.030	.842	2.031	−.033	.034	−.008
.748	2.024	−.038	.047	.004	.819	2.041	.015	.077	.039
.517	1.679	−.039	.007	−.013	.515	1.513	−.008	−.054	−.013
.353	1.392	−.086	−.088	−.082	.495	1.502	.014	−.042	.006
.236	1.251	−.057	−.105	−.074	.489	1.486	.009	−.051	−.001
.149	1.270	.087	.000	.052	.367	1.288	.010	−.112	−.017
.148	1.296	.115	.027	.079	.240	1.147	.076	−.122	.027
Mean		.057	.053	.043	Mean		.040	.070	.026

* *B—refers to results obtained in brass cell.*
S—refers to results obtained in steel cell.

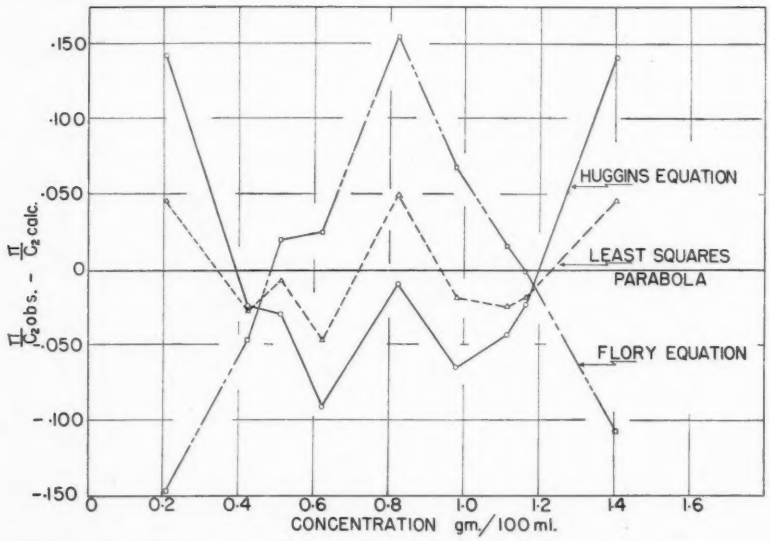

FIG. 3. *Deviations of experimental osmotic data from theoretical equations.*

Flory or the Flory–Huggins equation. Further evidence for this conclusion is shown in Fig. 3 which is a graph of deviation of the experimental value of π/C_2 from each calculated equation plotted against concentration. The data for Gelva V360 in chloroform have been chosen as representative. It is evident that the deviations are random about the unrestricted equation but follow a distinct trend in both the other cases.

Rather than base conclusions on this evidence, it was decided to make a careful statistical analysis of the data to determine whether these apparent differences were indeed significant. As an arbitrary criterion for rejection of data it was decided to include all values of π/C_2 whose deviation from the unrestricted quadratic equation was less than three times the standard deviation for all values of π/C_2 in the same solvent. On this basis it was not possible to reject any data even though certain values are open to considerable suspicion.

The results of the statistical analysis may be briefly summarized. The standard error was calculated for the intercept on the π/C_2 axis for each unrestricted quadratic equation. A "*t*-test" was then used to determine whether the calculated intercept differed significantly from the common intercept obtained by visual extrapolation of the curves (0.735 for Gelva V360-V360B and 0.875 for Gelva V360A). In three solvents (Gelva V360 in methyl ethyl ketone, Gelva V360B in ethyl acetate, and Gelva V360A in benzene (steel cell)) there was a significant difference in the intercepts at the 5% level but in no case was there a significant difference at the 1% level. However, a similar analysis showed that the intercepts predicted by the Flory and the Flory–Huggins equations were almost invariably significantly different from the intercepts predicted by the unrestricted parabola and hence were probably different from the common intercept extrapolated visually.

Equations for the visually-fitted curves were determined following the procedure of Bawn, Freeman, and Kamaliddin (1) and the dispersions of the experimental values about these lines were compared by means of an "*F*-test" with the dispersions about the least squares parabolas. The visually-fitted equations were found to be not significantly different from the simple least squares equations at the 1% level of significance. A similar test was used to compare the simple parabola with the Flory and the Flory–Huggins equations. The evidence is not conclusive but indicates a somewhat lower dispersion about the unrestricted parabola.

Discussion

1. An osmotic cell has been developed whose operation is reliable and whose results show good precision. For any one solvent, the average deviation of experimental values of π/C_2 about a simple quadratic equation fitted by the method of least squares is approximately ± 0.03 cm. This would correspond to an uncertainty of 4% in the molecular weights measured in this test. In different solvents, however, the intercepts obtained by extrapolating these least squares equations cover a range of ± 0.06. This uncertainty is apparently

consistent with the experimental precision and does not indicate a significant dependence of molecular weight on solvent. It appears, therefore, that the uncertainty in molecular weight is about 10% even though the experimental measurements are precise to ± 0.03 cm.

2. Caustic-swollen "Cellophane" membranes are suitable for use with polyvinyl acetate solutions.

3. Available theoretical equations do not describe the π/C_2 vs. C_2 curve in the concentration range usually employed and consequently are not satisfactory in predicting the value of the π/C_2 intercept at $C_2 = 0$.

It should be noted that the method of curve-fitting suggested by Flory for his recent equation is a visual one involving a log–log plot which will tend to obscure the differences between his equation and the simple empirical equation. Further experimental data may well prove that Flory's equation is required to account for the curvature of the π/C_2 vs. C_2 plot in the extremely dilute region but at present it seems to be inferior to the unrestricted quadratic equation.

4. A simple quadratic equation, whose constants are evaluated empirically, is adequate in describing the curvature of the π/C_2 vs. C_2 curve and predicts a common intercept for different solvents consistent with experimental error.

5. A smooth curve fitted visually to the experimental data is equally satisfactory in representing the curvature and in predicting the value of the intercept. Bawn has described a convenient method of analyzing such a curve.

6. There seems to be no effect of solvent on the value of the π/C_2 intercept in the polyvinyl acetate systems reported here.

Acknowledgment

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References

1. BAWN, C. E. H., FREEMAN, R. F. J., and KAMALIDDIN, A. R. Trans. Faraday Soc. 46: 862. 1950.
2. BROWNING, G. V. and FERRY, J. D. J. Chem. Phys. 17: 1107. 1949.
3. FLORY, P. J. J. Chem. Phys. 10: 51. 1942.
4. FLORY, P. J. J. Chem. Phys. 17: 1347. 1949.
5. HUGGINS, M. L. Ann. N.Y. Acad. Sci. 41: 1. 1942.
6. MASSON, C. R. and MELVILLE, H. W. J. Polymer Sci. 4: 337. 1949.
7. MILLER, A. R. Proc. Cambridge Phil. Soc. 39: 54. 1943.
8. ROBERTSON, R. E., MCINTOSH, R., and GRUMMITT, W. E. Can. J. Research, B, 24: 150. 1946.
9. SIRIANNI, A. F., WISE, L. M., and MCINTOSH, R. L. Can. J. Research, B, 25: 301. 1947.
10. TIMMERMANS, J. Physico-chemical constants of pure organic compounds. Elsevier Publishing Co. Inc., Amsterdam and New York. 1950.

A COMPARISON OF THE PROTECTIVE COLLOID INDEX AND POLAROGRAPH WAVE FOR NORMAL AND CANCER SERA[1]

By L. A. Munro and D. J. Boyd

Abstract

The protective colloid indices have been determined on the sera of 83 normal rats and on 74 rats with transplanted Walker carcinosarcoma. As previously found with humans, the sera from tumor animals have much less of the protective colloids than the sera of normals. When 0.12 ml. of serum was used, 82% of the cancerous animal gave residual dye of less than 51γ per ml., whereas 100% of the normals gave concentrations above this value. Half of the false negative tests were from rats having tumors less than 15 gm. in weight. When 0.10 ml. of serum was used, 89.2% had an index below 15γ with an average of 7.8γ while the index of 91% of the normals was above 15γ with an average of approximately 42γ. The protective indices and polarograms have been compared for the sera of 43 cancerous and 47 normal rats. In 91% of the cancerous animals characteristic curves were obtained having a wave height greater than 50 mm., with 4.2% false positives in the normal group. The two tests are of approximately equal value in classifying the sera. The serum constituents responsible for the polargraph wave are different from those indicated by the protective colloid index.

Protective Colloids in Cancer

In 1944 a study was made of the protective colloid index of the blood sera of cancer patients and normal individuals (6). The protective index of a lyophilic colloid is defined by the residual concentration of a standard sol in micrograms per milliliter after its treatment with an electrolyte in the presence of a given amount of the protecting colloid, under standard conditions. Cancer serum was found to give a much lower index than normal serum. Seventy of seventy-eight cancer patients (approximately 90%) gave values less than 15γ per ml. when 0.075 ml. of serum was used as compared with 3 from 70 normal individuals (approximately 4%). When a 0.10 ml. portion of serum was used in each test, the frequency – colloid index curves were very different, 86% of the normals gave values greater than 100γ per ml. whereas 80% of the cancer patients were below this range. The cancer group included a wide variety of cancer types in all stages of development.

The present work was undertaken to see whether transplanted tumors in animals would bring about a similar lower protective colloid index, and, if so, to establish the quantitative values for normal and tumor animals.

Methods

The colloid used in the test was Congo red purified by recrystallization from hot aqueous ethyl alcohol (10 gm. of dye + 150 ml. 95% ethyl alcohol + 150 ml. water). The mixture was filtered while hot through a Buchner using No. 1 Whatman paper, and the filtrate allowed to crystallize. The crystallized pro-

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duct was removed by filtration, washed with cold 95% ethyl alcohol, and the procedures repeated until the maximum adsorption was obtained with the Beckman quartz spectrophotometer.

The coagulating electrolyte was U.S.P. grade quinine monohydrochloride. Other electrolytes could be used, but this was found in the previous study to be the most satisfactory. Aluminum salts cannot be employed owing to the color changes produced in the dye at the pH of the hydrolyzed salt.

The rats were of the Wistar strain. The tumor, Walker carcinosarcoma 256, was implanted subcutaneously by trocar method into the groin of the animal. Blood samples were obtained by decapitation.

Procedure

The technique of the test is exacting. The glassware must be scrupulously clean. The adsorption of chromic ions from cleaning solution was found to influence the test and consequently a sulphuric acid – nitric acid mixture was substituted. Accurate pipetting is necessary since as little as 0.01 ml. of serum has a very great effect on the result. In subsequent experiments the serum was diluted and an equivalent amount used.

The chosen amount of serum was transferred to each of three conical centrifuge tubes containing 5 ml. of 0.10% Congo red. Five milliliter portions of a 0.30% solution of quinine monohydrochloride were then pipetted into separate tubes. At zero minus two minutes the electrolyte was dumped into the first tube and the whole mixed by pouring back and forth four times. This method of mixing was found to be better than shaking the tubes. At zero minus one minute the electrolyte was dumped into the second tube and at zero time the third tube was mixed. All tubes were then placed in a constant-temperature bath at 30°C.

After 30 min. in the bath, the tubes were removed and centifuged at 2400 r.p.m. for four minutes. The color in the supernatant was then evaluated in a Klett photelometer using a blue filter No. 42, from a standard curve for Congo red.

In the present study, the results reported are a mean of from two–five determinations depending on the amounts of sera available and the agreement obtained.

Preliminary trials, using human cancer and normal sera, were made. The marked differences reported by one of us were again observed. This indicated that the reagents and technique were satisfactory for the present work.

Results

P.C. Index

The results, using 0.10 ml. of serum for 74 cancer and 83 normal rats, are summarized in Table I and Fig. 1.

TABLE I

PROTECTIVE COLLOID INDEX USING 0.10 ML. SERUM

Residual dye, γ/ml.	Normals		Cancer	
	No. in group	Per cent of total	No. in group	Per cent of total
0–5	0	0	37	50.0
6–10	4	4.82	23	31.1
11–15	5	6.04	6	8.1
16–20	8	9.64	3	4.0
21–25	7	8.44	3	4.0
26–30	3	3.62	1	1.3
31–35	4	4.82	1	1.3
36–40	6	7.23	0	0
41–45	5	6.04	0	0
46–50	2	2.41	0	0
> 50	39	46.8	0	0
Total	83	99.9	74	99.8

The values for residual dye are given in micrograms (γ) of Congo red per milliliter.

Table II gives the results when 0.12 ml. of serum was used. The distribution curves are shown in Fig. 2.

It will be noted that the distribution curves are very different for cancer and normal sera. Using 0.10 ml. of serum, no cancerous animal, no matter how small the tumor, gave residual dye greater than 35γ. Only two of the 74 cancer animals gave a protective colloid index of greater than 25γ. If 15γ is taken as the dividing index between the protective value of cancer and normal sera as was done with humans, 89.3% of all cancer animals gave final dye concentrations below this figure. Four of the eight cancer sera which showed higher

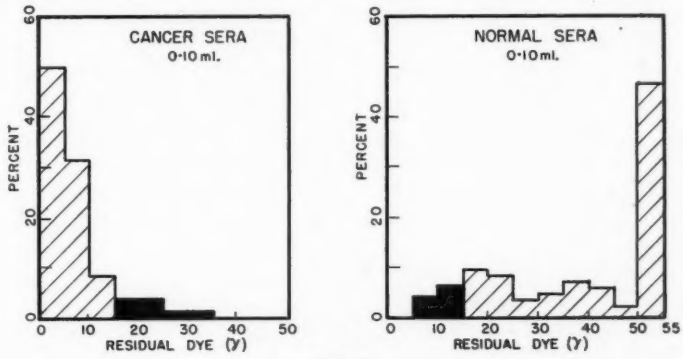

FIG. 1

FIG. 1. *Distribution curves for residual dye using 0.10 ml. of serum from cancerous and normal animals.*

TABLE II

PROTECTIVE COLLOID INDEX USING 0.12 ML. SERUM

Residual dye, γ/ml.	Normals		Cancer	
	No. in group	Per cent of total	No. in group	Per cent of total
0–5	0	0	8	14.8
6–10	0	0	17	31.5
11–15	0	0	10	18.5
16–20	0	0	4	7.4
21–25	0	0	3	5.5
26–30	0	0	1	1.9
31–50	0	0	1	1.9
> 50	61	100	10	18.5
Total	61	100	54	100

protective value had small tumors (less than 15 gm.). In the 70 animals with tumors greater than 15 gm., 94% had a protective colloid index below 15γ. On the other hand, 89% of the normal animals gave residual dye greater than 15γ per ml. when 0.10 ml. of sera was used. The average concentration of residual dye was 7.8γ per ml. for the cancer animals and approximately 42γ per ml. for the normals.

Examination of the results obtained for 0.12 ml. of serum reveals that with well developed tumors (tumors of the order of 30 gm. or more), the cancer sera were much less effective as protective agents than the normal sera. Indeed 100% of the "non-takes" gave values for the residual dye of at least 50γ per ml., i.e. there were no false positives, while in the case of the advanced tumor rats, only five of the 48 (10.6%), gave values of 50γ per ml.

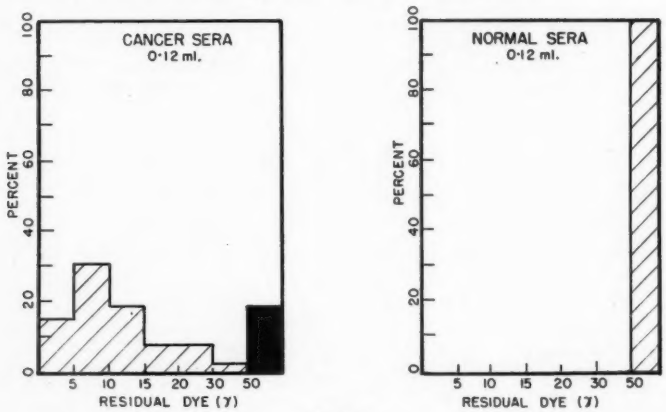

FIG. 2

FIG. 2. *Distribution curves for residual dye using 0.12 ml. of serum from cancerous and normal animals.*

Polarograph Studies

It seemed of interest to obtain the polarograms on the sera of rats bearing the Walker carcinosarcoma 256, and to compare the relative accuracy of the protective colloid index and the polarographic waves in indicating malignancy using the same animals, especially since the former involves the whole serum and the latter a specific fraction of the serum.

The polarograph has been used extensively to indicate differences between normal humans and cancerous individuals. According to the comprehensive survey of Winzler and Burk (10) 93% of some fourteen hundred cancer patients gave polarograph waves significantly higher than normal. There are very few papers in the literature dealing with polarographic studies in cancerous rats. Klatt *et al.* (3, 4) have studied the sera of tumor bearing rats and mice. They report a difference in the height in the waves for the cancerous and normal animals.

Winzler and Burk used three different tumors; the Jensen sarcoma, hepatoma 31, and a chemically induced primary hepatoma. They noted a marked increase in the serum proteose as indicated by the polarogram, with each of the above tumors. The sera of animals with hepatoma 31 were found to give a good correlation between tumor size and wave height until a maximum was reached about 15 days after implantation.

Methods

Polarograms were made using a technique similar to that of Waldschmidt-Leitz (9). By this method, 0.5 ml. of serum was pipetted into a centrifuge tube and the serum proteins were then precipitated using 1 ml. of a 25% solution of 5-sulphosalicylic acid. One milliliter of distilled water was next added and the mixture stirred. After centrifuging for 10 min. at 2400 r.p.m. the supernatant was poured into a No. 50 Whatman filter paper and allowed to drain. (By centrifuging first, the filtration was greatly accelerated.) An aliquot of the filtrate, 0.5 ml., was then measured into a polarograph vessel, containing 4 ml. of cobalt buffer solution and 8 ml. of distilled water.

A stream of nitrogen, which had been passed over heated copper shavings (450°C.) to remove traces of oxygen, was next allowed to bubble through the solution for approximately 10 min. The dropping cathode was then attached to the vessel and the cell connected to the instrument—a model XX Sargent Polarograph.

The polarograms were then made using a span potential of 2.0 v. In each case the bridge was initially set at about 0.6 v., since the double wave was completely recorded in this range (0.6–2.0). This was the only part of the curve in which we were interested. A sensitivity setting of 2–55 on this instrument was used for all polarograms. This sensitivity proved to be the most satisfactory, since it allowed good magnification of the second wave without the

pen running off the chart paper. By using the same sensitivity every time, the wave heights all became directly comparable.

Polarographic Results

The heights of the polarographic second waves were measured as indicated in the accompanying figure (Fig. 3). These polarograms are typical ones obtained from normals and from tumor bearing animals. Not only is the height of the wave greater with cancer serum but the appearance of the wave is different.

TABLE III

Cancer animals			Normal animals		
Wave height, mm.	Number	%	Wave height, mm.	Number	%
11–20	0	0	11–20	5	10.6
21–30	1	2.3	21–30	18	38.3
31–40	0	0	31–40	18	38.3
41–50	3	6.9	41–50	4	8.5
51–60	3	6.9	51–60	1	2.1
61–70	12	27.9	61–70	1	2.1
71–80	9	20.9	71–80	0	0
81–90	10	23.3	81–90	0	0
91–100	4	9.3	91–100	0	0
101–110	1	2.3	101–110	0	0
Total	43	99.8		47	99.9

The summarized data for 43 tumor rats and 47 healthy animals are given in Table III. The average height of the double wave with tumor serum is 77 mm. as compared with 31 mm. obtained with normal serum.

These results are embodied in the distribution curves given in Fig. 4.

Comparison and Discussion

A study of Table III and Fig. 4 reveals that in the majority of cases of well developed tumors, the polarographic double wave is higher than in normal animals. Thus in the 43 cancer sera examined polarographically, 39 (90.7%) gave wave heights greater than 50 mm. Five of the normals gave wave heights between 40–60 mm. It appears that the sera which gave waves less than 50 mm. are to be classified as normals. Those from 40–60 mm. might be classified as doubtful, while those producing wave heights greater than 60 mm. are definitely pathological.

Tables IV and V contain values obtained by both tests when made on the same sera samples for 41 cancer animals and 39 normals. A correct diagnosis is indicated by a + sign. Conversely, a − sign indicates a negative or incorrect finding. '*D*' indicates a doubtful diagnosis.

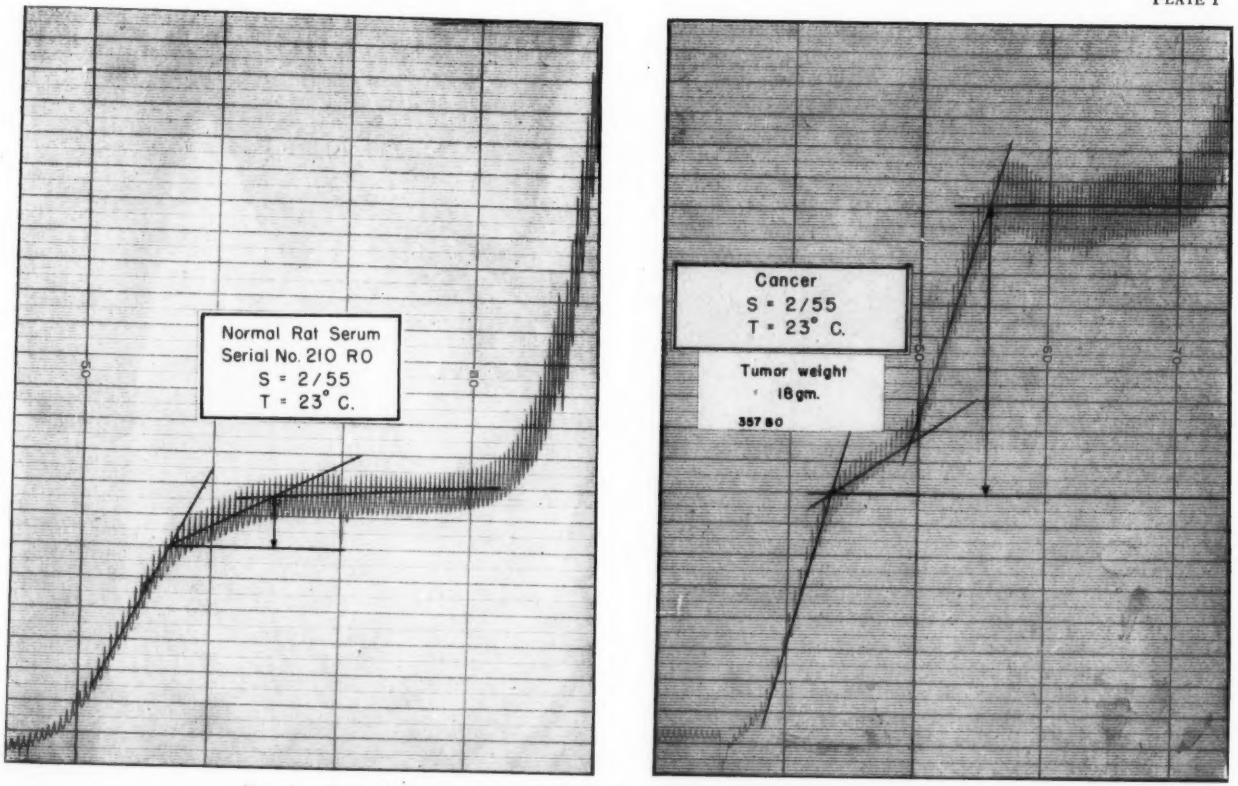

FIG. 3. *Typical polarograms for: (a) normal rat serum; (b) cancerous rat serum.*

With the protective colloid index, values under 15γ for 0.10 ml. portions are considered as being indicative of cancer. Animals giving values of 15–20γ are in a doubtful range unless the 0.12 ml. is below the ± group, in which case it would be considered as normal if the second tube is in the + group or cancerous if it is below 50γ.

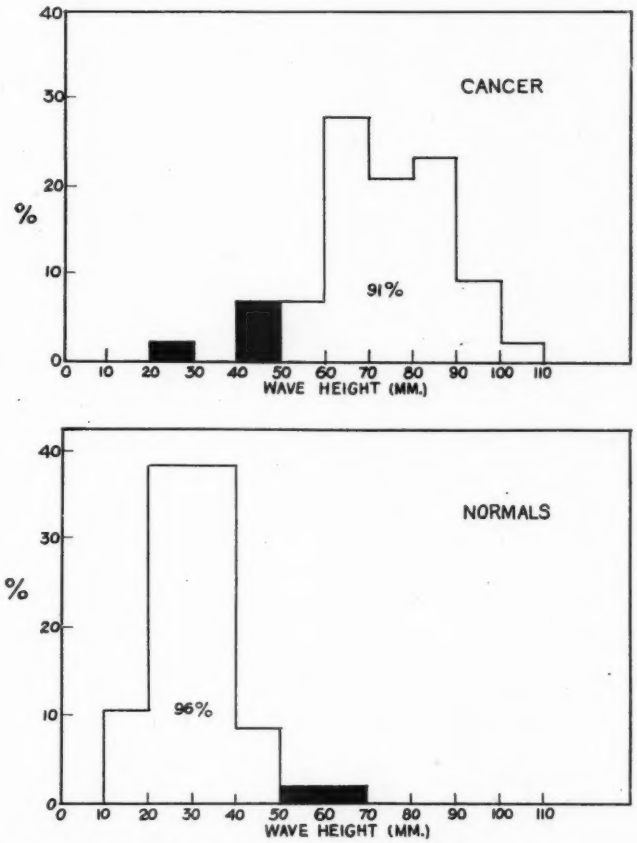

FIG. 4. *Distribution curves for wave heights of cancer and normal rat sera.*

The results embodied in Tables IV and V are summarized in Table VI.

The cancer sera diagnosed as 'doubtful' by both tests are from small tumor animals. An insufficient number of these cases has been examined to permit any final conclusions on the relative merits of the tests in detecting early stages of the disease. More work on this aspect of the problem is therefore necessary.

The differentiation of the cancer rats into advanced, medium, and small tumor cases is only approximate and rather arbitrary. The body weight of the animals may have a bearing on the problem. Thus, a tumor of 10 gm. may have

TABLE IV

CANCER ANIMALS

No.	Wave height, mm.	Polarograph diagnosis	Residual dye, γ/ml. (0.10 ml.)	Residual dye, γ/ml. (0.12 ml.)	P.C. Index diagnosis	Tumor weight
1	69	+	2.9	11.8	+	71
2	70	+	21.2	±	−	54
3	93	+	2.8	10.0	+	83
4	86	+	6.0	17.2	+	56
5	86	+	5.8	11.8	+	133
6	82	+	2.4	5.1	+	108
7	81	+	2.8	4.0	+	71
8	83	+	0.8	0.8	+	107
9	76	+	3.2	8.1	+	60
10	76	+	2.5	2.5	+	127
11	83	+	1.1	3.0	+	110
12	82	+	3.8	7.1	+	72
13	72	+	5.4	10	+	67
14	65	+	5.4	±	+	77
15	68	+	2.9	12.2	+	81
16	65	+	3.0	11.0	+	54
17	76	+	1.4	1.6	+	81
18	68	+	1.4	1.4	+	80
19	75	+	16.8	21.2	+	87
20	89	+	2.8	2.8	+	45
21	68	+	2.2	4.8	+	76
22	61	+	5.4	14.8	+	46
23	53	+	4.9	12.8	+	43
24	65	+	5.8	11.2	+	71
25	84	+	3.8	6.0	+	55
26	67	+	19.8	20	+	90
27	80	+	4.8	9.8	+	109
28	70	+	7.1	13.2	+	58
29	70	+	3.8	22	+	91
30	96	+	4.4	10.0	+	87
31	91	+	4.4	5.4	+	84
32	84	+	4.0	4.0	+	54
33	52	+	6.8	18.0	+	70
34	75	+	7.4	±	+	20
35	48	*D*	21.8	±	−	5
36	53	+	25	−	−	10
37	93	+	10	±	+	20
38	76	+	5.4	±	+	25
39	47	*D*	12.4	±	+	10
40	24	−	17	±	*D*	10
41	46	*D*	39	±	−	14

Note: ± indicates practically no coagulation: residual dye > 50 γ per ml.

more effect on a 125 gm. animal than on one weighing 230 gm. Pertinent data is not at hand. However, it appears that, when the tumors have attained a size of about 40 gm., the body weight of the animal is not a significant factor as regards changes in the blood serum constituents.

TABLE V

NORMAL ANIMALS

Stock No.	Wave height, mm.	Polarograph diagnosis	Residual dye, γ/ml. (0.10 ml.)	Residual dye, γ/ml. (0.12 ml.)	P.C. Index diagnosis
1	17	+	±	±	+
2	26	+	±	±	+
3	27	+	±	±	+
4	22	+	35	±	+
5	16	+	31	±	+
6	22	+	±	±	+
7	26	+	±	±	+
8	28	+	15.2	±	*D*
9	31	+	24.2	±	+
10	63	−	±	±	+
11	37	+	±	±	+
12	46	*D*	±	±	+
13	37	+	15.2	±	*D*
14	21	+	30.2	±	+
15	32	+	±	±	+
16	29	+	±	±	+
17	35	+	41.2	±	+
18	25	+	±	±	+
19	36	+	14.8	±	−
20	30	+	±	±	+
21	25	+	±	±	+
22	35	+	±	±	+
23	26	+	±	±	+
24	44	*D*	±	±	+
25	46	*D*	±	±	+
26	33	+	±	±	+
27	33	+	20.0	±	+
28	43	*D*	22.8	±	+
29	24	+	36.2	±	+
30	32	+	24.8	±	+
31	35	+	±	±	+
32	29	+	2.12	±	+
33	35	+	±	±	+
34	40	*D*	±	±	+
35	29	+	±	±	+
36	18	+	±	±	+
37	28	+	±	±	+
38	38	+	±	±	+
39	22	+	9.8	±	−

Note: ± indicates practically no coagulation: residual dye > 50 γ per ml.

These experiments appear to indicate that the polarograph and P.C. Index are of an approximately equal value when the 256 Walker carcinosarcoma is used.

TABLE VI

	Polarograph				P.C. Index			
	Positive	Negative	Doubtful	Total	Positive	Negative	Doubtful	Total
Cancer	37	1	3	41	36	4	1	41
Normal	33	1	5	39	35	2	2	39

It is certain that the particular serum constituents responsible for the polarograph test are not the same as those involved in the protective colloid test. The former is considered (3, 4) to be determined by and a measure of the proteoses of molecular weight 10,000 or so, containing—S-S—and S-H-linkages. It has been suggested that excessive proteose arises in cancer from the degradation of albumin possibly through the action of a proteolytic enzyme of Aberhalden. Others believe it arises through failure of the liver to metabolize normal amounts of polypetides or from impaired kidney functions.

Obviously since there is more proteose present in cancer and less of the protective colloids, the proteose cannot be the source of the protection of the Congo red against the electrolyte.

The exact nature of the changes indicated by the P.C. Index have not been determined. The most obvious suggestion is that the test depends on the decreased amount of serum albumin which is significantly low in cancer. The decrease in albumin is accompanied by an increase in α_1 and α_2 globulins and, in plasma, of fibrinogen (8).

Mider, Alling, and Morton (5) in their recent paper on plasma proteins in malignancy state that the correlation factor for decreased albumin and increased globulin indicated a high degree of variation. It was found in the previous work with human sera (6) that the A/G ratio was much less significant in indicating malignancy than the Protective Colloid Index, and that the correlation between the A/G and P.C. Index was poor.

Heubner and Jacobs (2) and also Reitstotter (7) from studies of the gold numbers of serum fractions, concluded that the euglobulin had a higher protective action than the albumin and other fractions.

Bernsohn and Borman (1) believe that one or more β-globulins and possibly α globulins are responsible for the protective action of spinal fluid on the gold sol in the Lange test, whereas gamma globulin has a coagulating action. Serum albumin, they state, is inert. We have found that the so-called albumin fraction, obtained by precipitation of the globulins with half saturated ammonium sulphate, after dialysis was approximately equal in its protective action to the whole serum on the basis of nitrogen content.

Tiselius (8) has shown the heterogeneous nature of such 'salted' fractions. It therefore seems probable that the decrease in protective colloid index in cancer is due, not to the lower concentration of albumin only, but to the conversion of albumin, euglobulin, and possibly other serum constituents, to much less active species.

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References

1. BERNSOHN, JOSEPH and BORMAN, EARL K. J. Clin. Invest. 26: 1026. 1947.
2. HEUBNER, W. and JACOBS, F. Biochem. Z. 58: 352. 1913; Chem. Abstracts, 8: 1802. 1914.
3. KLATT, T., RUSCH, H. P., DIRKSEN, A., and MELOCHE, V. W. Trans. Electrochem. Soc. 79: 413. 1941.
4. KOLTHOFF, I. M. and LINGANE, J. J. Polarography. Chap. 31. Interscience Publishers Inc. New York. 1941.
5. MIDER, G. B., ALLING, E. L., and MORTON, JOHN J. Cancer, 3: 56. 1950.
6. MUNRO, L. A. J. Phys. Chem. 48: 187. 1944.
7. REITSTOTTER, JOSEF.* Kolloid-Z. 28: 20. 1921.
8. TISELIUS, A. Biochem. J. 31: 1464. 1937.
9. WALDSCHMIDT-LEITZ, E. Angew. Chem. 51: 324. 1938.
10. WINZLER, R. J. and BURK, D. J. Nat. Cancer Inst. 4: 417. 1944.

** This author is correctly cited by T. Svedberg in Colloid Chemistry p. 231, Chem. Catalogue Co., but the author's abstract states 'Der geringste Goldzahl besitzen die Albumin, der grosste die Euglobuline . . .', although in the text of his paper (p. 22 and Table II), it is clear that the abstract should read the 'reciprocal of the gold number'. Unfortunately Chemical Abstracts repeats the error in the author's summary.*

CONTENTS OF VOLUME 29

ADAMS, G. A. and CASTAGNE, A. E.—Hemicelluloses of wheat straw, 109.

BAER, E. and NEWCOMBE, A. G.—I. Mesyl esters of glycerol, 805. Asymmetrically substituted mixed acid triglycerides. Acyl derivatives of L-α-monomesylin and D-α, β-dimesylin, 812.

BAIN, O. and OUELLET, C.—A dilution effect in reactions between dipolar molecules in solution, 646.

BAKER, B. E.—See Henneberry, G. O.

BAKER, C. A. N.—See Taylor, E. G.

BARNWELL, J. L.—See Watson, R. W.

BEAUBIEN, P. E.—See Eastham, A. M.

BERMAN, L., MEEN, R. H., and WRIGHT, G. F—The nitrolysis of hexamethylenetetramine VII. Intermediates in the nitrolysis, 767.

BERNSTEIN, H. J.—The calculation of the change in vibrational frequency due to changes in mass, geometry, and potential constants of a molecule, 284.

BETTS, R. H. and MACKENZIE, A. N.—Isotopic exchange reactions in the system bromine–bromate–hypobromous acid, 655. Formation and stability of hypobromous acid in perchloric acid solutions of bromine and bromate ions, 666.

BLADES, H. and WINKLER, C. A.—The reaction of nitrogen atoms with methane and ethane, 1022.

BLAKE, R. I.—See Van Cleave, A. B.

BLUNDELL, M. J.—See Stone, J. E.

BOIVIN, J. L.—See Gagnon, P. E.; Picard, J. P.

BOIVIN, J. L. and BOIVIN, P. A.—Preparation of N-substituted ureas from nitrosomethylureas, 478.

BOIVIN, P. A.—See Boivin, J. L.; Gagnon, P. E.

BOURNS, A. N.—See Lindsay, J. G.

BOWDEN, K. and MARION, L.—The biogenesis of alkaloids. IV. The formation of gramine from tryptophan in barley, 1037. The biogenesis of alkaloids. V. Radioautographs of barley leaves fed with tryptophan -β-C^{13}, 1043.

BOYD, D. J.—See Munro, L. A.

BRICE, C.—See Lemieux, R. U.

BROOK, A. G. and Wright, G. F—The significance of the "mercurinium ion" in oxymercuration, 308.

BROWN, W. H.—See Robinson, J. R.

BRUNET, P. E.—See McKay, A. F.

BRYCE, J. R. G.—See McKay, A. F.

CASTAGNE, A. E.—See Adams, G. A.

CAULE, E. J. and STEACIE, E. W. R.—The photolysis of mercury dipropyl, 103.

CHALMERS, B.—See Winegard, W. C.

CHARANDUK, R.—See Lemieux, R. U.

CHRISTIAN, W. R., GOGEK, C. J., and PURVES, C. B.—Studies in the polyoxyphenol series. V. Preparation from pyrogallol of the isomeric cyclohexane-1,2,3-triols and -1,2-diols, and of several isopropylidene and monomethyl derivatives, 911.

CHRISTIAN, W. R. and PURVES, C. B.—Relative stabilities of the isomeric cyclohexane-1,2,3-triol trinitrates and 1,2-diol dinitrates, 926.

COCKBURN, W. F.—A semimicro filtration apparatus, 715.

COCKBURN, W. F. and MARION, L.—The papilionaceous alkaloids. VIII. The structure of thermopsine, 13.

COOPE, J. A. R.—See Watson, R. W.

COX, R. H.—See Linburg, R. G.

COX, R. H. and SPENCER, E. Y.—The estimation of some 17-alkyl substituted steroids with the zinc chloride – acetyl chloride (Tshugaev) reagent and a postulated reaction mechanism, 217. The ultraviolet absorption spectra of several dehydration and oxidation derivatives of cholesterol, 398.

CRAIG, B. M.—See Youngs, C. G.
CRAIG, H. M.—See Gagnon, P. E.
CROCKER, I. H.—See Graham, R. P.
CVETANOVIĆ, R. J. and LE ROY, D. J.—Contribution to the theory of diffusion flames, 597.
DARWENT, B. DEB.—See Eastham, A. M.
DIAPER, D. G. M., KIRKWOOD, S., and MARION, L.—The biogenesis of alkaloids. III. A study of hyoscyamine biosynthesis using isotopic putrescine, 964.
DOUGLAS, B.—See Marion, L.
EASTHAM, A. M., DARWENT, B. DEB.—The chemistry of ethylene oxide. III. Reaction of ethylene oxide in amine solution, 585.
EASTHAM, A. M., DARWENT, B. DEB., and BEAUBIEN, P. E.—The chemistry of ethylene oxide. II. The kinetics of the reaction of ethylene oxide with amines in aqueous solution, 575.
EDMISTON, W. E. and WIESNER, K.—Synthesis of some phenanthridine derivatives, 105.
EMBREE, W. H., MITCHELL, J. M., and WILLIAMS, H. L.—Compositional heterogeneity of butadiene-acrylonitrile copolymers prepared in emulsion at 5°C., 253.
EPSTEIN, S. and Winkler, C. A.—Studies on RDX and related compounds. VI. The homogeneous hydrolysis of cyclotrimethylenetrinitramine (RDX) and cyclotetramethylenetetranitramine (HMX) in aqueous acetone, and its application to analysis of HMX in RDX (B), 731.
EVANS, H. G. V.—See Hodgson, G. W.
FAINER, P.—The reduction of nitroglycerin with titanous sulphate, 46.
FAIRBAIRN, D. and HARPUR, R. P.—The distillation, extraction, and chromatographic separation of the C_2–C_8 fatty acids, 633.
FARNAND, R.—See Stainsby, G.
FITCH, F. T. and RUSSELL, D. S.—The separation of the rare earths by ion-exchange procedures, 363.
FOLEY, W. T. and GIGUÈRE, P. A.—Hydrogen peroxide and its analogues. II. Phase equilibrium in the system hydrogen peroxide–water, 123. Hydrogen peroxide and its analogues. IV. Some thermal properties of hydrogen peroxide, 895.
FOLLOWS, A. G.—See Taylor, E. G.
FROST, G. B., MOON, K. A., and TOMPKINS, E. H.—The role of amorphous intermediate products in the dehydration of certain hydrated salts, 604.
FUNT, B. L. and MASON, S. G.—Influence of shear on the dielectric constant of liquids, 848.
GAGNON, P. E., BOIVIN, J. L., BOIVIN, P. A., and JONES, R. N.—The synthesis and ultraviolet spectra of some pyrazolones, 182.
GAGNON, P. E., BOIVIN, J. L., and GIGUERE, J.—Contribution to the study of 4-monosubstituted-3-amino-5-pyrazolones, 328. Synthesis of 5-substituted 1-anilino-4-iminobarbituric acids, 1028.
GAGNON, P. E., BOIVIN, P. A., and CRAIG, H. M.—Synthesis of amino acids from substituted cyanoacetic esters, 70.
GAGNON, P. E., NOLIN, B., and JONES, R. M.—Synthesis and ultraviolet absorption of some pyrazolones, 843.
GAUDRY, R.—The synthesis of amino acids from 2,3-dihydrofuran DL-ornithine, DL-proline, and DL-α-amino-δ-hydroxyvaleric acid, 544.
GAUDRY, R. and McIVOR, R. A.—Synthèse des acides aminés à partir de la rhodanine, 427.
GAUVIN, W.—See Wiebe, A. K.
GIGUERE, J.—See Gagnon, P. E.; Lemieux, R. U.
GIGUÈRE, P. A.—See Foley, W. T.; Phibbs, M. K.
GIGUÈRE, P. A. and LAMONTAGNE, D.—Polarographic determination of benzoyl peroxide and cumene hydroperoxide, 54.
GILLESPIE, T. and LANGSTROTH, G. O.—The effect of relative humidity on coagulation and surface losses of ammonium chloride smoke, 133. The aging of ammonium chloride smokes, 201.
GILLIES, A., WILLIAMS, H. L., and WINKLER, C. A.—Studies of RDX and related compounds. III. The reaction to form RDX from ammonium nitrate and formaldehyde in acetic anhydride, 377.

GOGEK, C. J.—See Christian, W. R.
GOGEK, C. J., MOIR, R. Y., MCRAE, J. A., and PURVES, C. B.—An improved synthesis of *cis-cis-trans*-cyclohexane-1,2,3-triol, 938.
GOGEK, C. J., MOIR, R. Y., and PURVES, C. B.—The benzoate and the methyl ether of 4-hydroxycyclohexene, 946.
GRAHAM, R. P., MACNAMARA, J., CROCKER, I. H., and MACFARLANE, R. B.—The isotopic constitution of germanium, 89.
GRIFFITHS, F. H.—See McRae, J. A.
HABGOOD, T. E.—See McRae, J. A.
HARDWICK, T. J. and ROBERTSON, E.—Ionic species in ceric perchlorate solutions, 818. Association of ceric ions with sulphate (a spectral study), 828.
HARPUR, R. P.—See Fairbairn, D.
HARWOOD, V. D.—Cupriethylenediamine as a reagent for hemicellulose separation from wheat straw holocellulose, 974.
HASKINS, R. H.—See Lemieux, R. U.
HATTON, W. G.—See McKay, A. F.
HEAL, H. G.—The influence of cation radius and salt concentration in the salting out of ethyl ether from aqueous nitrate solutions at 25°C., 563.
HENNEBERRY, G. O., OLIVER, W. F., and BAKER, B. E.—Synthesis of C^{14} carboxyl-labeled phenylalanine, 229.
HEYDING, R. D. and WINKLER, C. A.—Solvent effect on iodide exchange, 790.
HODGSON, G. W., EVANS, H. G. V., and WINKLER, C. A.—The exchange of iodine between inorganic iodides and *n*-butyl iodide, 60.
HOLLIES, N. R. S. and MCINTOSH, R. L.—The dielectric constant of disulphur decafluoride, 494.
HOLMES, O. G —See McCallum, K. J.
INGRAM, A. R.—Electronic interpretation of the uncatalyzed reaction between resorcinol and formaldehyde in alcohols and dioxane, 863.
JAILLET, J. B. and OUELLET, C.—Cinétique de l'oxydation de la formaldéhyde par l'eau oxygenée en solution alcaline, 1046.
JONES, R. A. and WINKLER, C. A.—Reactions in dissociated water vapor, 1010.
JONES, R. N.—See Gagnon, P. E.
KATES, M. and MARION, L.—Gelsemine. III. Reduction with lithium aluminum hydride, 37.
KATZ, M., WILSON, L. G., and RIBERDY, R.—The oxidation of carbon monoxide by solid silver permanganate reagents. IV. Chemical composition in relation to activity, 1059.
KELLY, R. B., WHITTINGHAM, D. J., and WIESNER, K.—The structure of ryanodine. I., 905.
KIRKWOOD, S.—See Diaper, D. G. M.
KIRKWOOD, S. and MARION, L.—The biogenesis of alkaloids. II. The origin of the methyl groups of hordenine and choline, 30.
KRISTJANSON, A. M. and WINKLER, C. A.—Exchange reactions of iodide ion with aromatic iodides, 154.
LAMONTAGNE, D.—See Giguère, P. A.
LANGSTROTH, G. O.—See Gillespie, T.
LAVIGNE, R.—See Marion, L.
LEITCH, L. C. and MORSE, A. T.—Synthesis of *cis*- and *trans*-1,2-dichloroethylene-*d*, 1034.
LEMAY, L.—See Marion, L.
LEMIEUX, R. U.—The biochemistry of the Ustilaginales. III. The degradation products and proof of the chemical heterogeneity of ustilagic acid, 415. The mercaptolysis of glucose and galactose pentaacetates, 1079.
LEMIEUX, R. U. and CHARANDUK, R.—Biochemistry of the Ustilaginales. VI. The acyl groups of ustilagic acid, 759.
LEMIEUX, R. U. and GIGUERE, J.—Biochemistry of the Ustilaginales. IV. The configurations of some β-hydroxyacids and the bioreduction of β-ketoacids, 678.
LEMIEUX, R. U., THORN, J. A., BRICE, C., and HASKINS, R. H.—Biochemistry of the Ustilaginales. II. Isolation and partial characterization of ustilagic acid, 409.

LEONARD, N. J.—See Marion, L.
LEROY, D. J.—See Cvetanović, R. J.; Osberg, G. L.
LINBURG, R. G., COX, R. H., and SPENCER, E. Y.—Some observations on the copper oxide oxidation of cholesterol, 716.
LINDSAY, J. G., BOURNS, A. N., and THODE, H. G.—C^{13} isotope effect in the decarboxylation of normal malonic acid, 192.
LUSENA, C. V.—Estimation of ribonucleic acid with orcinol, 107.
LYNE, M. L.—See Taylor, E. G.
MCCALLUM, K. J. and HOLMES, O. G.—Chemical effects of the Cl^{35} (γ,n) Cl^{34} reaction in sodium chlorate, 691.
MACCORMACK, K. E. and SCHNEIDER, W. G.—Isotherms of sulphur hexafluoride in the critical temperature region, 699.
MACFARLANE, R. B.—See Graham, R. P.
MACHUTCHIN, J. G.—See Ralph, A. O.
MCINTOSH, R.—See McLeod, L. A.
MCINTOSH, R. L.—See Hollies, N. R. S.; Trost, W. R.
MCIVOR, ROBERT A.—See Gaudry, R.
MCKAY, A. F., BRYCE, J. R. G., and RIVINGTON, D. E.—Preparation and nitration of 1-substituted-2-nitramino-2-imidazolines, 382.
MCKAY, A. F. and HATTON, W. G.—Aryldiguanidines, 718.
MCKAY, A. F., PICARD, J. P., and BRUNET, P. E.—The structures of nitroguanidine and its derivatives, 746.
MCKAY, A. F. and THOMAS, H. P.—Preparation of substituted nitroguanidines and the cyclization of N-γ-nitroxybutyl-N′-nitroguanidine, 391.
MACKENZIE, A. N.—See Betts, R. H.
MCLEOD, L. A. and MCINTOSH, R.—An evaluation of osmotic pressure measurements using polyvinyl acetate systems, 1104.
MACNAMARA, J.—See Graham, R. P.
MCRAE, J. A.—See Gogek, C. J.; Manske, R. H.
MCRAE, J. A., VANORDER, R. B., GRIFFITHS, F. H., and HABGOOD, T. E.—6-Iodometameconine and 3-iodometahemipinic acid, 482.
MAJKA, S.—See Winegard, W. C.
MALLARD, T. M.—See Youngs, C. G.
MANSKE, R. H., MCRAE, J. A., and MOIR, R. Y.—3-Bromometameconine, 526.
MARION, L.—The papilionaceous alkaloids. XIV. Concerning spathulatine and nonalupine, 959. See Bowden, K.; Cockburn, W. F.; Diaper, D. G. M.; Kates, M.; Kirkwood, S.
MARION, L. and DOUGLAS, B.—The papilionaceous alkaloids. XIII. Identity of octalupine with hydroxylupanine, 721.
MARION, L., LAVIGNE, R., and LEMAY, L.—The structure of sedamine, 347.
MARION, L. and LEONARD, N. J.—The papilionaceous alkaloids. XI. Identity of genisteine with *l*-α-isosparteine, 297. The synthesis of dehydrolupanines and the stereochemistry of the C_{15} lupin alkaloids, 355.
MARION, L., TURCOTTE, F., and OUELLET, J.—The papilionaceous alkaloids. IX. Natural occurrence of the alkaloids of the α-isosparteine group, 22.
MASON, S. G.—See Funt, B. L.
MEEN, R. H.—See Berman, L.
MILLER, D. M. and WINKLER, C. A.—The measurement of the absolute rates of removal of lead and tellurium mirrors by a free radical stream, 537.
MITCHELL, J. M.—See Embree, W. H.
MOIR, R. Y.—See Gogek, C. J.; Manske, R. H.
MOON, K. A.—See Frost, G. B.
MORSE, A. T.—See Leitch, L. C.
MOSES, G. B.—See Sirianni, A. F.
MOSES, G. B. and PUDDINGTON, I. E.—The rheological properties of some soap–oil systems, 996.

MUNRO, L. A. and BOYD, D. J.—A comparison of the protective colloid index and polarograph wave for normal and cancer sera, 1115.
NEISH, A. C.—Analysis of mixtures of simple aliphatic alcohols by partition chromatography, 552.
NEWCOMBE, A. G.—See Baer, E.
NICHOLLS, R. V. V.—See Van Straten, S. F.
NOLIN, B.—See Gagnon, P. E.
OLIVER, W. F.—See Henneberry, G. O.
ORR, R. J. and WILLIAMS, H. L.—The compositional heterogeneity of butadiene–styrene copolymers synthesized at − 18°C. in emulsion, 270. The role of alpha-ketols in the low-sugar redox recipe for low temperature emulsion copolymerization, 949.
OSBERG, G. L. and LE ROY, D. J.—The polymerization of styrene sensitized by acetyl radicals, 333.
OUELLET, C.—See Bain, O.; Jaillet, J. B.; Ouellet, L.
OUELLET, J.—See Marion, L.
OUELLET, L. and OUELLET, C.—Sur la flamme froide de l'ether, 76. Influence de gaz étrangers sur la flamme froide de l'ether, 986.
PHIBBS, M. K. and GIGUÈRE, P. A.—Hydrogen peroxide and its analogues. I. Density, refractive index, viscosity, and surface tension of deuterium peroxide – deuterium oxide solutions, 173. Hydrogen peroxide and its analogues. III. Absorption spectrum of hydrogen and deuterium peroxides in the near ultraviolet, 490.
PICARD, J. P.—See McKay, A. F.
PICARD, J. P. and BOIVIN, J. L.—Study of nitration of N,N′-disubstituted hydrazines, 223.
PUDDINGTON, I. E.—See Moses, G. B.; Sirianni, A. F.; Stainsby, G.
PURVES, C. B.—See Christian, W. R.; Gogek, C. J.
RAFF, R. A. V. and SILVERMAN, B. H.—Influence of alcohols upon the uncatalyzed reaction between resorcinol and formaldehyde, 857.
RALPH, A. O., MACHUTCHIN, J. G., and WINKLER, C. A.—Studies on RDX and related compounds. V. The effect of delayed addition of ammonium nitrate, and the rate of disappearance of hexamine, in the Bachmann reaction, 725.
RIBERDY, R.—See Katz, M.
RIPLEY, L. G. and WATSON, R. W.—Hydrolysis and polymerization of cyclic dimethylethylene oxalates, 970.
RIVINGTON, D. E.—See McKay, A. F.
ROBERTSON, E.—See Hardwick, T. J.
ROBINSON, J. R. and BROWN, W. H.—Diphenylcyanamide derivatives, 1069.
RUSSELL, D. S.—See Fitch, F. T.
SALLANS, H. R.—See Youngs, C. G.
SCHIFF, H. I. and STEACIE, E. W. R.—The reactions of H and D atoms with cyclic and paraffin hydrocarbons, 1.
SCHNEIDER, W. G.—Sound velocity and sound absorption in the critical temperature region, 243. See MacCormack, K. E.
SCOTT, E. J. Y. and STEACIE, E. W. R.—The mercury photosensitized reactions of benzene at high temperatures, 233.
SHU, P.—See Thorn, J. A.
SILVERMAN, B. H.—See Raff, R. A. V.
SIRIANNI, A. F., MOSES, G. B., and PUDDINGTON, I. E.—Particle shape in thixotropic suspensions, 166.
SMITH, E. A.—See Zlatkis, A.
SPENCER, E. Y.—See Cox, R. H.; Linburg, R. G.
SPOONER, R. C. and WETMORE, F. E. W.—Molten salts. Electrical conductivity in the system silver chloride – silver nitrate, 777.
STAINSBY, G., FARNAND, R., and PUDDINGTON, I. E.—The preparation of anhydrous sodium stearate, 838.
STEACIE, E. W. R.—See Caule, E. J.; Schiff, H. I.; Scott, E. J. Y.; Wijnen, M. H. J.

STONE, J. E., BLUNDELL, M. J., and TANNER, K. G.—The formation of lignin in wheat plants, 734.

SWENSON, E. G. and THORVALDSON, T.—The alcohol-glycerol method for the determination of free lime, 140.

TANNER, K. G.—See Stone, J. E.

TAYLOR, E. G., BAKER, C. A. N., and LYNE, L. M.—Conductance of ammonium nitrate in water – nitric acid – nitrogen pentoxide mixtures at various temperatures, 452.

TAYLOR, E. G. and FOLLOWS, A. G.—Some freezing point and conductance measurements in the system nitric acid – acetic acid, 461.

TAYLOR, E. G., LYNE, L. M., and FOLLOWS, A. G.—Conductance measurements in water – nitric acid – nitrogen pentoxide mixtures at various temperatures, 439.

THALL, B. M.—See Winegard, W. C.

THODE, H. G.—See Lindsay, J. G.

THOMAS, H. P.—See McKay, A. F.

THORN, J. A.—See Lemieux, R. U.

THORN, J. A. and SHU, P.—A new apparatus for rapid carbon determination by wet combustion, 558.

THORVALDSON, T.—See Swenson, E. G.

TOMPKINS, E. H.—See Frost, G. B.

TROST, W. R.—The addition compounds of silicon tetrachloride and amines, 877. The addition compounds of silicon tetrachloride with aliphatic amines, 1075.

TROST, W. R. and MCINTOSH, R. L.—The kinetics of the thermal decomposition of disulphur decafluoride, 508.

TURCOTTE, F.—See Marion, L.

TURNER, R. C. and WINKLER, C. A.—Dropping amalgam electrodes, 469.

VAN CLEAVE, A. B. and BLAKE, R. I.—The reaction of dioxane with hydrogen bromide, 785.

VANORDER, R. B.—See McRae, J. A.

VAN STRATEN, S. F., NICHOLLS, R. V. V., and WINKLER, C. A.—Exchange reactions of alicyclic iodides with sodium iodide, 372.

WATSON, R. W.—See Ripley, L. G.

WATSON, R. W., COOPE, J. A. R., and BARNWELL, J. L.—Physical constants and estimation of stereoisomeric 2,3-butanediols, 885.

WETMORE, F. E. W.—See Spooner, R. C.

WHITTINGHAM, D. J.—See Kelly, R. B.

WIEBE, A. K., GAUVIN, W., and WINKLER, C. A.—Hydrogen overvoltage and potential build-up at copper cathodes, 301.

WIESNER, K.—The structure of jaborandine, the alkaloid of *Piper jaborandi* Velloso. I., 352. See Edmiston, W. E.; Kelly, R. B.

WIJNEN, M. H. J. and STEACIE, E. W. R.—Photolysis of 2,2′,4,4′-tetradeuterodiethyl ketone, 1092.

WILLIAMS, H. L.—See Embree, W. H.; Gillies, A.; Orr, R. J.

WILLIAMS, H. L. and WINKLER, C. A.—Studies on RDX and related compounds. IV. The Bachmann or combination process, 642.

WILSON, L. G.—See Katz, M.

WINEGARD, W. C., MAJKA, S., THALL, B. M., and CHALMERS, B.—Eutectic solidification in metals, 320.

WINKLER, C. A.—See Blades, H.; Epstein, S.; Gillies, A.; Heyding, R. D.; Hodgson, G. W.; Jones, R. A.; Kristjanson, A. M.; Miller, D. M.; Ralph, A. O.; Turner, R. C.; Van Straten, S. F.; Wiebe, A. K.; Williams, H. L.

WRIGHT, G. F—See Berman, L.; Brook, A. G.

YOUNGS, C. G., MALLARD, T. M., CRAIG, B. M., and SALLANS, H. R.—Component fatty acids of rapeseed oil, 871.

ZLATKIS, A. and SMITH, E. A.—The synthesis of some stereoisomeric derivatives of decahydronaphthalene, 162.

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Contents

Page

The Biogenesis of Alkaloids. IV. The Formation of Gramine from Tryptophan in Barley—*K. Bowden and Léo Marion* - - - - 1037

The Biogenesis of Alkaloids. V. Radioautographs of Barley Leaves Fed with Tryptophan-β-C^{14}—*K. Bowden and Léo Marion* - - 1043

Cinétique de l'Oxydation de la Formaldéhyde par l'Eau Oxygénée en Solution Alcaline—*Jean-B. Jaillet et Cyrias Ouellet* - - - 1046

The Oxidation of Carbon Monoxide by Solid Silver Permanganate Reagents. IV. Chemical Composition in Relation to Activity —*Morris Katz, L. G. Wilson, and R. Riberdy* - - - - - - 1059

Diphenylcyanamide Derivatives—*J. R. Robinson, and W. H. Brown* 1069

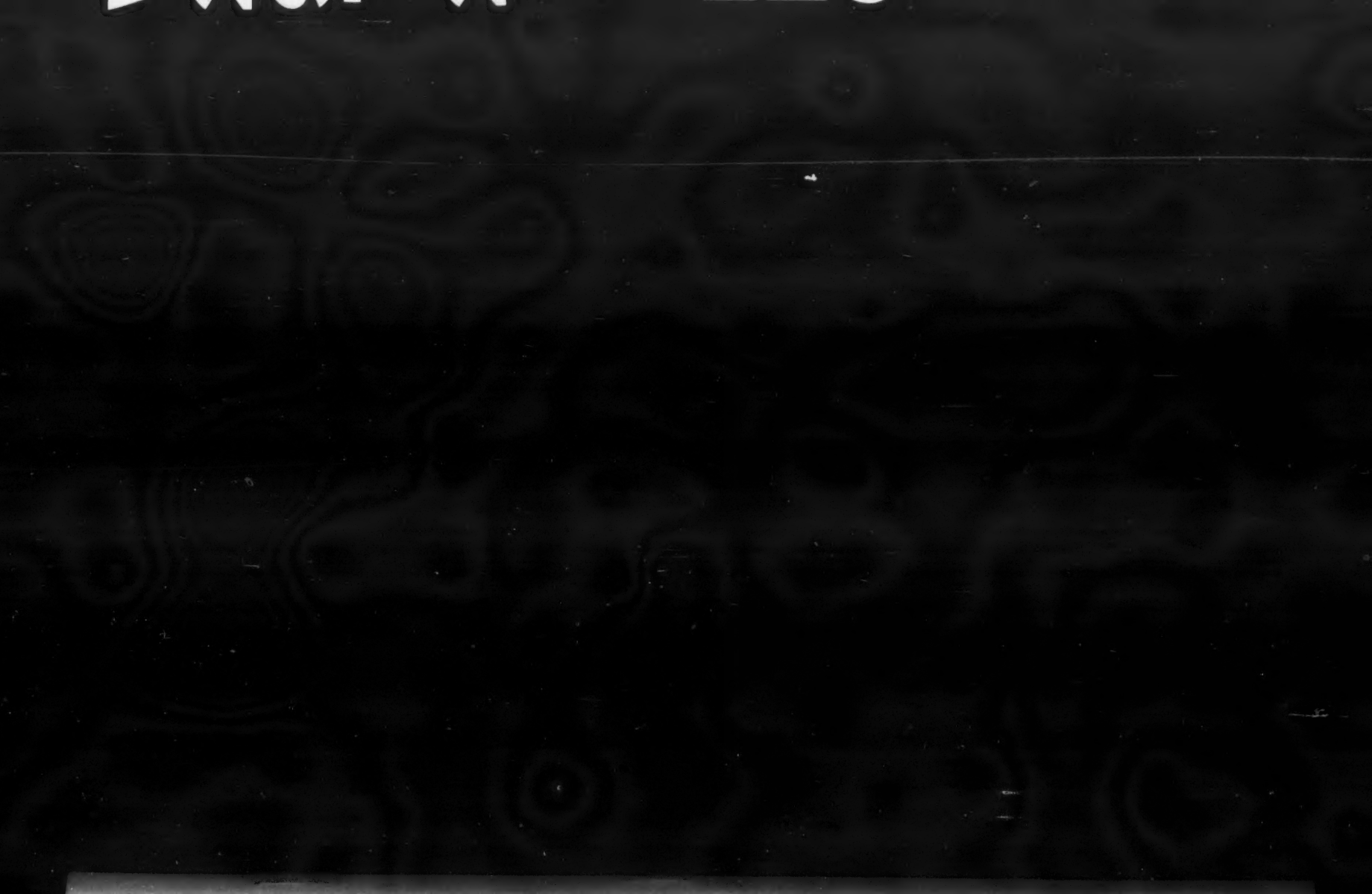
The Addition Compounds of Silicon Tetrachloride with Aliphatic Amines—*W. R. Trost* - - - - - - - - - - - - - - - 1075

The Mercaptolysis of Glucose and Galactose Pentaacetates— *R. U. Lemieux* - - - - - - - - - - - - - - - - - - 1079

Photolysis of 2, 2′, 4, 4′-Tetradeuterodiethyl Ketone—*M. H. J. Wijnen and E. W. R. Steacie* - - - - - - - - - - - - 1092

An Evaluation of Osmotic Pressure Measurements Using Polyvinyl Acetate Systems—*L. A. McLeod and R. McIntosh* - - - - 1104

A Comparison of the Protective Colloid Index and Polarograph Wave for Normal and Cancer Sera—*L. A. Munro and D. J. Boyd* 1115

Contents of Volume 29 - - - - - - - - - - - - - - - - - 1126

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